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Contents of previous issues of the JOURNAL OF ANIMAL SCIENCE are indexed in the Agricultural Index which is available in most libraries and in the November issue of each complete volume of the JOURNAL.

ANTIBIOTIC SUPPLEMENTS IN RATIONS FOR GROWING AND FATTENING LAMBS [1, 2, 3]

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ALTHOUGH the use of certain antibiotics in the rations of poultry and swine is generally accepted, the value of antibiotics in the rations of ruminants is less well established. The results of most experiments reviewed by Reid *et al.* (1954) have shown that small amounts of aureomycin or terramycin added to the rations of young calves resulted in increased gains in weight. However, with older cattle the results of aureomycin supplementation have varied from a favorable influence, as indicated by gains of yearling steers on a high roughage ration (Perry *et al.*, 1954), to a depression of the apparent digestibility of crude fiber and decreased nitrogen retention in steers (Bell *et al.*, 1951; and Horn *et al.*, 1953 respectively). Neumann *et al.* (1951) were unable to obtain any benefit by supplementing the rations of yearling heifers with aureomycin.

Likewise the results of experiments in which the effects of antibiotic supplementation in lamb feeding were studied have been variable and inconsistent. Colby *et al.* (1950a) reported that young lambs which received 9 mg. aureomycin, supplied with an APF supplement, per pound of concentrate (the first two weeks they received 18 mg. aureomycin per lb. of concentrate) gained less than their controls both before and after weaning. Moreover after weaning the feed intake of lambs in the lot receiving the ration supplemented with aureomycin was lower than that of the lambs in the control lot.

Definite detrimental effects were observed in fattening lambs by Colby *et al.* (1950b) when 100 mg. aureomycin with or without all

[1] Published with the approval of the Illinois Agricultural Experiment Station.
[2] The authors wish to acknowledge the assistance of William Gaither and B. C. Breidenstein, Department of Animal Science in collecting data and for grading the lamb carcasses, in trials II and III, respectively; Dr. N. D. Levine, College of Veterinary Medicine, for parasite study; and Garvey Hayden and Richard Levis of Armour and Company, Chicago, in obtaining carcass data.
[3] The aureomycin HCl and aurofac were supplied through the courtesy of R. F. Elliott, Lederle Laboratories, American Cyanamid Company, Pearl River, New York. The TM-5 and P-2 supplements were supplied through the courtesy of W. K. Warden, Pfizer and Company, Terre Haute, Indiana.
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known B-vitamins were given daily by capsule. The lambs receiving the supplement lost weight while their controls gained more than 0.5 lb. daily. Lambs which received 100 mg. penicillin daily went off-feed, lost weight, and had diarrhea for almost a week; and lambs which were given 100 mg. streptomycin daily gained slightly less than their controls.

Jordan and Bell (1951) were able to obtain favorable results by supplementing the ration of both suckling and fattening lambs with 5 to 15 mg. aureomycin per head daily. All lambs given the aureomycin appeared normal and had a higher rate of gain than their controls. The two lots of feeder lambs receiving the aureomycin required 22 and 20 percent, respectively, less concentrates per unit of gain than the lambs on an unsupplemented ration. In four subsequent trials these investigators (1954) reported that the average daily gains of suckling lambs which received from 4.3 to 10.8 mg. aureomycin per head daily were 0.04 lb. less and 0.08, 0.09, and 0.10 lb. more than their controls, respectively.

Kinsman and Riddell (1952) reported that the rate of gain in lambs was not affected by supplementing a creep ration with 15 mg. aureomycin, terramycin, or penicillin per lb. of ration.

Jordan (1952) fed feeder lambs 7.2, 10.8, and 14.4 mg. aureomycin daily without causing them to go off feed or increasing the incidence of scouring. Lambs in one lot which received 7.2 mg. aureomycin supplementation failed to gain as rapidly as their controls, while lambs in another lot which received 7.2 mg. aureomycin supplementation and lambs in the lot that obtained 10.8 mg. aureomycin supplementation gained more rapidly than their controls. Lambs in two lots which were fed rations supplemented with 14.4 mg. aureomycin daily gained less and required more feed per unit gain than did their controls.

Luce *et al.* (1953) observed that lambs which received 10 mg. aureomycin daily for two or three weeks after birth gained approximately the same as the controls, and lambs that received 10 mg. aureomycin daily from one or three weeks of age to weaning gained less than the controls.

In three experiments Bridges *et al.* (1953) found that lambs which received 1.1, 2.2, 3.2, 4.3, 5.0, or 15.0 mg. aureomycin per lb. of fattening ration performed as well or better than their controls. These investigators reported that there was an apparent improvement in feed efficiency in lambs fed rations which contained from 2.2 to 5.0 mg. aureomycin supplementation per lb. ration.

In a nitrogen balance study with feeder lambs Tillman and MacVicar (1954) found that 10 mg. crystalline aureomycin daily did not affect significantly the amount of nitrogen retained or the digestibility of the ration.

Turner and Hodgetts (1952) demonstrated that a single dose of 750 mg. aureomycin administered by mouth to adult Merino sheep had a very depressing effect upon ruminal digestion which was followed by diminished appetite and loss of weight.

Experimental

Three feeding trials involving 190 suckling and feeder lambs were conducted to study the influence of rations supplemented with antibiotics on growth rate, feed efficiency, and carcass grade.

The first trial was conducted in the spring and summer of 1952 with 50 Shropshire-western crossbred ewe and wether lambs. The lambs ranged from 11 to 40 (av. 28.2) days in age and varied from 15 to 38 (av. 22.8) lb. in weight. The lambs with their dams were divided into two lots as uniformly as possible with regard to twins or singles, sex, weight, and age of the lambs. The lambs were kept in adjoining dry lots (part of each lot was sheltered) during the entire trial. They were creep fed alfalfa hay and a concentrate mixture consisting of 30 percent ground corn, 30 percent whole oats, 30 percent wheat bran, and 10 percent soybean meal; and had free access to water and trace mineralized salt. The concentrate mixture fed to lambs in lot 2 was supplemented with 10 mg. aureomycin HCl per lb. of concentrate. The ewes were fed twice daily in open feeding troughs. Lambs in both lots had equal opportunity to eat feed with the ewes, but the amount of feed consumed in that way appeared to be negligible. After the loss of one lamb in each lot (diagnosed as enterotoxemia) all remaining lambs were vaccinated with 5 cc. Clostridium perfringens type D bacterin and treated with 5 cc. Clostridium perfringens type D antitoxin. On the 86th day of the test the ewes were removed from both lots. The lambs were weighed on the 119th day which was used as the final weight although they were shipped to market four days later. The individual carcass weight and "warm" carcass grades were obtained at the time of slaughter.

The second trial was started in the fall of 1952 with 30 western black-faced feeder lambs weighing an average of 62.2 lb. The lambs were gate cut into two adjoining dry lots and appeared to be comparable with respect to size, type, etc. All lambs were vaccinated with

Clostridium perfringens type D bacterin before the trial began. A ground corn, soybean meal, chopped alfalfa hay mixed ration containing 50 percent hay at the beginning and 40 percent hay at the end of the trial was self-fed. The ration of lot 2 was supplemented with 5.5 mg. aureomycin HCl per lb. of ration. On the 93rd day and on the 100th day of the trial the five heaviest lambs, and on the 107th day the five remaining lambs in each lot, were weighed, shorn, slaughtered, and carcass data collected.

The third trial started in May, 1953, with 111 late spring Hampshire-western and Shropshire-western crossbred ewe and wether lambs. The age and sex of the lamb, breed of sire, and whether single or twin were considered in dividing the lambs into six adjoining dry lots. One-half of the lambs in each lot was vaccinated with Clostridium perfringens type D bacterin. Alfalfa hay and a concentrate mixture containing 88.5 percent corn-and-cob meal, 10 percent soybean meal, 1 percent steamed bone meal, and 0.5 percent trace mineralized salt were fed in a creep to all lots. In order to insure uniformity of the ration the corn-and-cob meal was made with 80 percent ground shelled corn and 20 percent ground cobs. The ewes were on pasture during the day and were kept in the lots with the lambs during the night until the lambs were weaned on the 42nd day of the test. The lambs in lot 1 served as the controls. The rations of the lambs in the remaining lots were supplemented as follows: lot 2, crystalline aureomycin HCl; lot 3, aurofac containing 4.02 gm. aureomycin per lb.; lot 4, TM-5 containing 5.3 gm. terramycin per lb.; lot 5, P-2 containing 2.3 gm. diamine penicillin per lb.; and, lot 6, aurofac, TM-5, and P-2 in ratio to furnish equal parts of the three antibiotics. The supplementation was at a rate to furnish 10 mg. of antibiotic per lb. of concentrate mixture. On the 70th day of the trial, due to machinery breakdown, finely ground corn had to be substituted for coarsely ground corn which had been used in the ration. On the 99th day of the trial all lambs which weighed 75 pounds or more were shipped to a meat packing company where individual carcass weights and grades were obtained. Lamb carcasses in trials II and III were graded by the same grader.

Results and Discussion

The data on growth rates and feed efficiencies are summarized in table 1, and the carcass data are shown in tables 2 and 3.

Trial I

The results in table 1 are averages for only those lambs that survived the test and were slaughtered. Two control lambs died in lot 1, one apparently from enterotoxemia and the other from an unknown cause. In lot 2 in which the ration that contained aureomycin HCl was fed two lambs died from enterotoxemia, one lamb died from an undetermined cause, and one lamb was crippled in shipping to slaughter. Also three sick lambs in lot 2 recovered after treatment with 10 cc. Clostridium perfringens type D antitoxin. These results show that aureomycin HCl at the level used in this experiment did not eliminate enterotoxemia. The average growth rate of the lambs which received the ration containing the aureomycin HCl was higher, almost reaching the 5 percent significance level, and they required 19 percent less feed per unit of gain than did the control lambs. Since lambs in both lots were group fed, the feed consumed by the two lambs lost in lot 1 and the feed consumed by the four lambs lost in lot 2 was charged to the surviving lambs of each lot. The average of the "warm" carcass grades shown in table 2, graded on the kill floor of a meat packing company, was higher for the lambs (P less than .01, assigning equally spaced scores to the several grades) which received the aureomycin HCl supplemented ration. In addition to the average weight gains the average dressing percents favored the lambs receiving the ration containing aureomycin HCl. The dressing percents were calculated from the individual carcass weights and the corresponding individual final weights obtained prior to shipping to market. Thus, the dressing percent absorbed a 7 percent shrinkage in weight which occurred during shipping.

The increased dressing percent in favor of the lambs that were fed the ration which contained the aureomycin HCl may have been due to the increased degree of finish indicated by the relative carcass grades. Apparently parasites were not an important factor in this test as the fecal egg counts were very low and were approximately the same in the two lots.

Trial II

These rations, self-fed, gave satisfactory average performances. One lamb (lot 2) which was off-feed for a few days during the second month of the test yielded the carcass with the lowest grade. There was no

TABLE 1. SUMMARY OF GROWTH AND FEED EFFICIENCY DATA

Trial	I		II		III					
Kind of lambs	Suckling		Feeder		Suckling					
Lot No.	1	2	1	2	1	2	3	4	5	6
No. of lambs	26	24	15	15	18	18[1]	20	18	18	18
Antibiotic[2]	None	..	None	..	None	..	..	..	..	..
Aureomycin HCl, mg.	..	7.2	..	5.5	..	7.6	..	..	..	..
Aurofac, mg.	..	..	..	..	..	..	7.4	..	..	2.5
TM-5, mg.	..	..	..	..	..	..	..	7.4	..	2.5
P-2, mg.	..	..	..	..	..	..	..	..	7.5	2.5
Deaths[3]	2	4	0	0	0	0	0	1	0	0
Initial weight, lb.	22.6	23.0	61.1	63.4	31.5	30.4	32.6	29.2	30.4	32.1
Final weight, lb.	59.7	65.6	98.7	104.1	64.5	69.0	69.6	64.3	66.3	66.3
Gain, lb.	37.1	42.6	37.6	40.7	33.0	38.6	37.0	35.1	35.9	34.2
Days on test[4]	119	119	100	100	70	70	70	70	70	70
Av. daily gain	.31	.36	.38	.41	.47	.55	.53	.50	.51	.49
99-day										
Weight,[5] lb.					75.7	81.6	80.8	77.6	78.1	79.1
Gain, lb.					44.2	51.2	48.2	48.4	47.7	47.0
Av. daily gain, lb.					.45	.52	.49	.49	.48	.47
Feed per lb. gain										
Concentrate	3.8	3.1	4.3	3.9	2.6	2.5	2.4	2.6	2.5	2.6
Roughage	1.5	1.2	3.6	3.3	.9	.8	.9	.9	.8	.9
Total	5.3	4.3	7.9	7.2	3.5	3.3	3.3	3.5	3.3	3.5

[1] Lot 2, trial III, started with 19 lambs but one lamb which refused to eat was removed.
[2] Mg. antibiotic per pound of total ration consumed.
[3] These lambs are excluded from the remaining data.
[4] Trial II, 1/3 lambs fed 93; 1/3 lambs fed 100; and 1/3 lambs fed 107 days.
[5] Trial III was for 99 days; physical character of ration changed on 70th day.

difference in the average fleece weights or the average dressing percentages of the two lots. The lambs receiving the ration supplemented with 5.5 mg. aureomycin per lb. of total ration had a higher average daily gain, almost reaching the 5 percent significance level; higher average carcass grades ($P = 0.12$, on the basis of equal spacing of the several grades), and required 8 percent less feed per unit of gain.

TABLE 2. SUMMARIZED CARCASS DATA OF LAMBS IN TRIAL I

Lot No.	Treatment	Final Experimental Weight-lb.	Dressing Percent[1]	Carcass Weight-lb.	Carcass Grades			
					Choice	Good	Utility	Cull
1	Control	59.7	45.5	27.2	4	8	7	5
2	Aureomycin HCl	65.6	48.9	32.1	10	7	2	1

[1] Absorbs a 7 percent live weight shrinkage during shipping 135 miles to market.

Trial III

The average daily gains in all lots were considered good for dry lot summer feeding. One lamb (lot 4) died from unknown cause, and one lamb which refused to eat was removed (lot 2). Although hot weather may have been a contributing factor, the change in the physical form of the ration on the 70th day of the experiment is believed to have caused the decreased feed consumption which occurred after the feed change. It was observed that the lambs which were consuming the largest amounts of feed, and which were the fastest gaining lambs, appeared to be the ones most affected. It was several days before all lambs became adjusted to the changed ration and were back on full-feed. Both the 70- and the 99-day weights are given because of this feed change. The average gains were higher for the lambs receiving rations supplemented with antibiotics. Statistical analysis of the 70-day weights showed that the lambs in the six lots differed significantly at the 5 percent level. Lambs in lots 1 and 2 of trial III received rations similar to rations used in trials I and II. Hence the differences in average daily gain between the lambs on the control rations and the lambs on the rations supplemented with aureomycin HCl for the three trials were compared. They were found to agree and were pooled to give an average advantage for the aureomycin HCl supplemented ration of 0.055 ± 0.014 pound per day.

In view of the fact that in young calves, as well as in swine and poultry, certain antibiotics have stimulated growth, such responses

might be expected in lambs, particularly young lambs. Although several hypotheses have been suggested as to the role of antibiotics in animal feeding, their influences on certain microorganisms should not be regarded as an unimportant one. Existing data show that the incidence and severity of scouring in calves have been reduced by antibiotic supplementation. Whitehair (1954) has pointed out that much of the effect of antibiotic supplementation may be by depressing subclinical infections.

In these three trials, carried out on premises which had been continuously occupied by sheep for many years, the lambs that received antibiotic supplements gave higher average performances. Although sheep husbandry practices, climatic conditions, and other factors will affect the health conditions, it is very likely that the amounts and kinds of contaminations and infestations would differ between closely confined sheep and sheep under less restricted conditions such as the range. It should be emphasized that antibiotics are not a substitute for good management and feeding practices, but they may aid in a good program by reducing the number of "unthrifty" and "poor doing" lambs. In a farm flock program it would seem advisable to use every available aid in promoting an early rapid growth to market weight and finish in lambs if such aids are economical and do not injure the final product.

Table 3 shows that a higher percentage of lambs which received rations supplemented with antibiotics reached the shipping weight of 75 pounds at the end of trial III. Tables 2 and 3 also show that the carcass grades of the lambs which received the rations supplemented with aureomycin or terramycin were higher than those of lambs which received the control rations. Fast growing lambs not only make more economical gains but are ready for market before the hot summer months which are unfavorable lamb feeding months in many areas.

Under the conditions of these feeding trials aureomycin HCl added to growing-fattening lamb rations at levels from 5.5 to 7.6 mg. per lb. of ration improved the rate and efficiency of gain but did not eliminate losses from enterotoxemia. The improvement in gain and efficiency was sufficient to show that aureomycin HCl has a practical place in lamb rations. Further work is needed to establish what antibiotic, or combination of antibiotics, gives the best response and what level of supplementation is most beneficial with the various methods of feeding which are used in the different production and feeding areas.

TABLE 3. SUMMARIZED CARCASS DATA OF LAMBS IN TRIALS II AND III

Lot No.	Treatment	No. lambs slaughtered[1]	Percent of surviving lambs slaughtered[1]	Dressing percent[2]	Grades								
					Prime			Choice			Good		
					Top	Av.	Low	Top	Av.	Low	Top	Av.	Low
Feeder lambs													
1	None	15	100	55.9	–	–	3	3	7	2	–	–	–
2	Aureomycin HCl	15	100	56.0	–	1	4	6	3	–	1	–	–
Suckling lambs													
1	None	10	56	49.6	–	2	3	3	1	–	1	–	–
2	Aureomycin HCl	14	72	49.1	–	–	10	2	2	–	–	–	–
3	Aurofac	16	80	50.6	–	3	6	3	4	–	–	–	–
4	TM-5	10	59	49.8	1	–	5	4	–	–	–	–	–
5	P-2	12	67	47.7	–	–	5	5	1	–	–	1	–
6	Aurofac, TM-5, P-2	11	61	48.6	–	3	3	3	1	1	–	–	–

[1] Only lambs that had reached a live weight of 75 pounds or more at the end of the feeding trials were slaughtered.
[2] Trial II lambs slaughtered at the experiment station. Trial III lambs slaughtered at Chicago, and the dressing percentage included 8 percent average live weight shrinkage.

Summary

Three feeding trials involving 190 suckling and feeder lambs were conducted to study the influence of rations supplemented with antibiotics on growth rate, feed efficiency, and carcass grade.

Rations supplemented with aureomycin HCl gave small but consistently higher average daily gains in all of the trials. When the results of the three trials were pooled, the lambs which received rations containing the aureomycin HCl made average daily gains higher by 0.055 $\pm$ 0.014 pound.

The average feed efficiencies of the lambs which received the rations supplemented with aureomycin HCl were higher than the average feed efficiencies of the lambs which received the control rations.

The average carcass grades of the lambs which received rations that contained the aureomycin HCl were higher than the average grades of their controls. The difference was highly significant in trial I on the basis of equal spacing of the several grades.

The average performance of the lambs which received the ration supplemented with aurofac nearly paralleled the average performance of the lambs in the same trial which received the ration supplemented with aureomycin HCl.

The average performance of the lambs fed the rations supplemented with TM-5 or P-2 was approximately the same or slightly better than the average performance of their controls, but the differences were not significant.

In trial I 7.2 mg. aureomycin HCl per lb. of ration did not eliminate losses from enterotoxemia.

These results indicate that antibiotics have a practical use in growing and fattening rations for lambs by improving rate and efficiency of gains and by reducing the number of unthrifty lambs.

Literature Cited

Bell, M. C., C. K. Whitehair, and W. D. Gallup. 1951. The effect of aureomycin on digestion in steers. Proc. Soc. Exp. Biol. and Med. 76:284.

Bridges, J. H., J. C. Miller, W. G. Kammlade, Jr., and H. O. Kunkel. 1953. Effects of various levels of aureomycin in fattening lambs. JOURNAL OF ANIMAL SCIENCE 12:660.

Colby, R. W., F. A. Rau, and J. R. Couch. 1950a. Effect of feeding an 'animal protein factor' concentrate to young lambs. Am. Jour. Physiol. 163:418.

Colby, R. W., F. A. Rau, and J. C. Miller. 1950b. The effect of various antibiotics on fattening lambs. (abstract) JOURNAL OF ANIMAL SCIENCE 9:652.

Horn, L. H., Jr., R. R. Snapp, and L. S. Gall. 1953. A study of the effects of antibiotics upon the digestion of feed nutrients by yearling steers, with bacteriological data. (abstract) JOURNAL OF ANIMAL SCIENCE 12:921.

Jordan, R. M. 1952. Aureomycin supplements in lamb fattening rations. JOURNAL OF ANIMAL SCIENCE 11:566.

Jordan, R. M. and T. D. Bell. 1951. Effect of aureomycin on growing and fattening lambs. (abstract) JOURNAL OF ANIMAL SCIENCE 10:1051.

Jordan, R. M. and T. D. Bell. 1954. Effect of aureomycin supplements on suckling lambs. JOURNAL OF ANIMAL SCIENCE 13:450.

Kinsman, D. M. and W. H. Riddell. 1952. Antibiotic in creep ration of suckling lambs. (abstract) JOURNAL OF ANIMAL SCIENCE 11:769.

Luce, L. C., B. H. Schneider, E. D. Taysom, and E. E. Goodwin. 1953. Antibiotics for lambs. The National Wool Grower, vol. 43, no. 11, p. 17.

Neumann, A. L., R. R. Snapp, and L. S. Gall. 1951. The long-time effect of feeding aureomycin to fattening beef cattle, with bacteriological data. (abstract) JOURNAL OF ANIMAL SCIENCE 10:1058.

Perry, T. W., W. M. Beeson, E. C. Hornback, and M. T. Mohler. 1954. Aureomycin for growing and fattening beef animals. JOURNAL OF ANIMAL SCIENCE 13:3.

Reid, J. T., R. G. Warner, and J. K. Loosli. 1954. Antibiotics in the nutrition of ruminants. Agr. and Food Chem. 2:186.

Tillman, A. D. and R. W. MacVicar. 1954. The effect of aureomycin upon the digestibility of ration components. Okla. Misc. Pub. No. MP-34, p. 92.

Turner, A. W. and V. E. Hodgetts. 1952. Depression of ruminal digestion in adult sheep by aureomycin. Austr. Jour. Agr. Res. 3:453.

Whitehair, C. K. 1954. Antibiotics in livestock rations. Okla. Misc. Pub. No. MP-34, p. 74.

THE EFFECT OF ALFALFA ASH UPON THE DIGESTIBILITY AND UTILIZATION OF COTTONSEED HULLS BY SHEEP[1]

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BURROUGHS *et al.* (1950) found that the addition of alfalfa ash promoted a greater utilization by cattle of a ration containing large amounts of corn cobs. Klosterman *et al* (1953) fed a fattening ration containing corn and cob meal and poor quality timothy hay to steers, and found that the addition of alfalfa ash or cobalt improved the utilization of the ration. Chappel *et al* (1952) found that a "synthetic" alfalfa ash compounded from inorganic salts to resemble the chemical composition of alfalfa ash also promoted a greater utilization by sheep of a ration containing large amounts of corn cobs.

Cottonseed hulls are obtained as a major by-product of the cottonseed oil industry and are available in amounts sufficient for livestock feeding in a large area of the United States. As this product, like corn cobs, contains substantial amounts of substances resistant to digestion, it seemed desirable to study the effect of alfalfa ash upon its digestibility and utilization.

Experimental Procedure

Digestibility Trial

The basal ration (ration 1) was a semi-purified type in which nearly all the minerals except calcium, phosphorus, chlorine, sodium, and sulfur were supplied by cottonseed hulls. The composition of the basal ration and daily intake in gm. were as follows: cottonseed hulls, 360; cerelose (corn sugar), 184.8; mazola (corn oil), 14.4; urea, 12.0; di-calcium phosphate, 14.4; sodium sulfate, 1.9; gelatin, 9.6; sodium chloride, 2.4; and vitamin A-D feeding oil (3000 I U of A and 6000 I U of D/gm.), 0.5. Twenty-eight gm. of alfalfa ash were added to the basal ration daily to form the experimental ration (ration 2). The

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chemical composition of the cottonseed hulls and the basal ration is shown in table 1.

The concentrate portion of the ration was mixed in a Hobart mixer and stored in a cool place until used. The cottonseed hulls, which constituted sixty percent, and the concentrate portion, which constituted forty percent of the ration, were weighed separately and mixed at the time of feeding. The alfalfa ash, which was prepared by burning in an open tank, followed by further ashing in a muffle furnace at 600° C., was also added at the time of feeding.

Western wether lambs were the experimental animals. Two weeks prior to being placed on their respective rations each lamb was drenched

TABLE 1. CHEMICAL COMPOSITION OF FEEDS AND BASAL RATION

Feed	Dry Matter %	Crude Protein %	Ether Extract %	Crude Fiber %	NFE %	Ash %	Organic Matter %	Energy cal./gm.
Cottonseed Hulls	93.9	3.7	1.1	40.8	45.4	2.8	91.1	4037
Basal Ration	93.3	9.1	2.7	24.6	53.2	3.7	89.62	3810

with a preparation containing phenothiazine. Twelve lambs were randomly divided into two groups. One group received the basal ration while the other group received the basal diet, plus 28 gm. of alfalfa ash daily. Each group was placed in a large box stall equipped with individual feeding stanchions. The animals were fed in the stanchions twice daily, and when not eating were allowed the freedom of the stall with free access to water.

The lambs were kept on constant feed intake during a successive 10-day preliminary period and a 10-day collection period. Collection of feces was made by means of a light harness and bag similar to that described by Garrigus and Rusk (1939). The feces were collected daily and the total collection was dried for 24 hours in a forced-air oven maintained at 60° C. Individual collections were transferred to a large container and at the end of the collection period the entire collection was mixed, sampled, and sample ground for proximate chemical analysis. The feed and feces were chemically analyzed essentially by the methods of the AOAC (1950). The gross energy content was determined with an Emerson fuel calorimeter equipped with an adiabatic jacket. The data were analyzed statistically by the "t" test (Snedecor, 1946). Blood samples were taken from all sheep at the end of the digestion trial for red blood cell count and hemoglobin determination.

Depletion-Repletion Trials

It was noted during the digestibility trial that the sheep receiving the basal ration lost weight while the sheep receiving alfalfa ash gained weight on approximately the same amount of feed. Therefore, a depletion-repletion technique was designed to study the effect of various additives for "depleted" animals. The depletion phase consisted of feeding the basal ration for 28 days, followed by a repletion phase of 21 days during which the effect of the additive was studied. Weights were taken at weekly intervals during both phases.

First trial

Two western wether lambs were the experimental subjects. During the depletion phase they were individually-fed as much as they would consume of the basal diet. At the end of this phase one animal was given 28 gm. of alfalfa ash daily in addition to the basal diet while the other lamb continued receiving the basal diet.

Second trial

Three western wether lambs were the experimental subjects in this trial. The depletion-repletion regimen was again followed with alfalfa ash as the additive. At the end of the 21-day repletion period all animals were returned to the basal diet for 21 days.

Third trial

Nine western wether lambs were used as experimental animals, and at the end of the depletion phase was divided into 3 equal groups. One group received a daily supplement of cobalt sulfate (ration 1) at a level approximating the amount of cobalt in 28 grams of alfalfa ash while another group received 45 grams per day of synthetic alfalfa ash (ration 2) of the following composition in gm.: $KHCO_3$, 960; K_2HPO_4, 348; $CaCl_2$, 277; $Ca(OH)_2$, 348; $MgSO_4 \cdot 7H_2O$, 592; $FeSO_4 \cdot 7H_2O$, 500; $NaHCO_3$, 319; $Na_2B_4O_7 \cdot 10H_2O$, 5.7; $MnSO_4 \cdot H_2O$, 3.0; $CuSO_4 \cdot 5H_2O$, 5.0; ZnO, 1.06; $CoCl_2 \cdot 6H_2O$, 0.007; and MoO_3, 0.003. The third group received 28 gm. per day of alfalfa ash in addition to the basal ration. At the end of the repletion period the animals which had received either synthetic alfalfa ash or alfalfa ash were given the basal diet for 21 days.

Results and Discussion

Digestibility trial

The results of the digestibility trial are summarized in table 2.

The addition of alfalfa ash to the basal ration improved the apparent digestibility of each ration constituent. In the cases of dry matter, crude fiber, ether extract, nitrogen-free extract, organic matter and energy the differences between the treatments were highly significant (p of .01). In the case of crude protein the difference was less but nevertheless was significant (p of .05). The added alfalfa ash had the greatest effect

TABLE 2. THE EFFECT OF ALFALFA ASH UPON THE DIGESTIBILITY OF A RATION CONTAINING COTTON SEED HULLS

	Digestibility Coefficients						
Ration	Dry Matter %	Crude Protein %	Ether Extract %	Crude Fiber %	NFE %	Organic Matter %	Energy %
Basal	51.0	40.4	78.8	35.6	60.3	51.9	47.9
Basal plus Alfalfa Ash	59.6**	44.6*	86.0**	46.5**	69.9**	61.3**	57.0**

* Significant.
** Highly significant.

on the digestion of crude fiber as indicated by coefficients of 35.6 and 46.5 by the sheep receiving the basal ration and the alfalfa ash supplemented ration, respectively. Visual inspection of the dried feces revealed that the feces from animals receiving the control ration contained a noticeable amount of linters, whereas none could be seen in the feces obtained from the animals receiving alfalfa ash in their ration.

These results suggest that the added alfalfa ash supplied one or more inorganic elements which were low in the basal ration, thereby promoting more efficient digestibility. Other factors such as total ash, anion-cation balance, buffering effect, and balance of inorganic elements, particularly the trace minerals (Plumlee *et al.*, 1953), must be considered in rationalizing the effect of alfalfa ash on digestibility of some roughages.

The average hemoglobin values were 10.9 and 11.6 percent for the sheep receiving the basal and the basal plus alfalfa ash rations, respectively, and the red blood counts were 7,450,000 and 7,840,000 cells per cmm for the basal and basal plus alfalfa ash treated lambs, respectively.

Depletion-Repletion Studies

The results of the depletion-repletion studies are shown in table 3. Without exception, the sheep receiving the basal diet for 28 days lost weight. During the first 7 to 10 days after the animals were placed on the basal diet, their appetites were good and they exhibited no abnormal behavior conditions. After about one week to 10 days on the basal diet,

TABLE 3. THE EFFECT OF ALFALFA ASH, SYNTHETIC ALFALFA ASH AND COBALT UPON THE GAINS OF SHEEP RECEIVING A RATION CONTAINING COTTONSEED HULLS

	Average daily gains during				
		Repletion			
Trial and No. of Sheep	Depletion Basal Ration Lb.	Alfalfa Ash Lb.	Synthetic Alfalfa Ash Lb.	Cobalt Lb.	Depletion [1] Basal Ration Lb.
Trial 1					
1	—0.31				—0.24
1	—0.14	0.48			
Trial 2					
3	—0.25	0.40			—0.35
Trial 3					
3	—0.12	0.21			—0.35
3	—0.17		0.22		—0.44
3	—0.11			—0.24	

[1] Denotes depletion period which followed the repletion period except in trial 1.

the animals began to eat less feed and developed abnormal craving for other feeds as exhibited by their chewing on wood in the stalls and on the clothes of the attendant. During the last week of the depletion regimen the animals were actually eating the wool from each other.

When alfalfa ash was added to the rations of the depleted animals they improved in appearance, ceased to have depraved appetites, and showed weight gains by the end of the first week. Swift *et al.* (1952) in calorimetric studies showed that alfalfa ash increased the digestibility of the energy of corn cobs by sheep and that there was a significant increase in metabolizable energy despite an increase in the amount of methane. The results of this experiment show that the increased digestible energy obtained by supplementing the basal ration with alfalfa ash was also available to the host and indicates further that a part of this energy was stored in the body.

When synthetic alfalfa ash was added during the repletion period, the sheep responded as well as those receiving the natural alfalfa ash. This is in agreement with the results of Chappel *et al.* (1952) which showed that synthetic alfalfa ash was almost as effective as natural alfalfa ash in increasing the digestibility by sheep of a ration containing corn cobs.

Cobalt did not improve the basal ration. In fact, there is a suggestion that it had a depressing effect as evidenced by the lambs losing an average of 0.24 lb. per day when receiving cobalt and only 0.11 lb. per day when receiving the basal ration during the depletion phase.

Summary

The addition of alfalfa ash to a semi-purified ration for sheep in which all minerals except calcium, phosphorus, sodium, chlorine and sulfur were supplied by cottonseed hulls improved the digestibility of all ration components.

A marked loss in weight was noted in sheep receiving the basal ration, and it was found that this condition could be reversed by addition of alfalfa ash. A depletion-repletion regimen was therefore feasible, and was used to study the effect of various additives to the basal ration. With this technique, a synthetic alfalfa ash promoted weight gain equal to that on natural alfalfa ash, while cobalt resulted in no improvement.

Literature Cited

Burroughs, Wise, Paul Gerlaugh and R. M. Bethke, 1950. The influence of alfalfa ash and fractions of alfalfa hay upon the digestion of ground corn cobs. JOURNAL OF ANIMAL SCIENCE 9:207.

Chappel, C. F., R. J. Sirny, C. K. Whitehair and Robert MacVicar, 1952. Effect of minerals on digestion of low-quality roughage by sheep. JOURNAL OF ANIMAL SCIENCE 11:758.

Garrigus, W. P. and H. P. Rush, 1939. Some effects of the species and stage of of maturity of plants on the forage consumption of grazing steers. Ill. Agr. Expt. Sta. Bul. 454.

Klosterman, E. W., L. E. Kunkle, O. G. Bentley and W. Burroughs (1953). Supplements to poor quality hay for fattening cattle. Ohio Agric. Expt. Sta. Res. Bul. 732.

Plumlee, M. P., Robert Totusek and W. M. Beeson. 1953. The effect of adding trace minerals to rations of identical twin beef cattle. JOURNAL OF ANIMAL SCIENCE 12:928.

Swift, R. W., R. L. Cowan, G. P. Barron, K. H. Maddy and E. C. Grosse. 1951. The effect of alfalfa ash upon roughage digestion in sheep. JOURNAL OF ANIMAL SCIENCE 10:434.

EFFECT OF RATION ON VITAMIN SYNTHESIS IN RUMEN OF SHEEP [1]

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THE disposal of rumen contents from packing plants constitutes, in many instances, a major waste-disposal problem. If an economically feasible way could be found to utilize this material, the disposal problem would be solved and the packing industry would have an additional by-product. Since it is known that the vitamin concentration in rumen contents varies greatly depending upon the type and quality of ration fed, information was needed on the specific effect of ration variation on the vitamin content of the rumen contents. It might be feasible to up-grade the material sufficiently by increasing the vitamin content to justify processing. This study was therefore instituted with the aim of measuring the effect of variation of the quality of roughage, the type of readily-available carbohydrate and the mineral constituents of the ration on the synthesis of three representative B-complex vitamins, namely, riboflavin, nicotinic acid and pantothenic acid.

Experimental

Experimental animals

Sixteen grade western yearling wethers ranging in weight from 70 to 90 lb. were used in this experiment. The lambs were assigned at random to their respective rations, two animals on each ration.

Management of animals

All lambs were fed in individual stanchions to assure controlled intake. When not eating, they were allowed the freedom of a large stall, free of bedding. Water was provided *ad libitum*. Rations fed are shown in table 1. Rations 2 through 6 were fed in sufficient quantity to provide a total of 800 gm. of air-dry material per day. Animals would consume only 600 gm. of the unsupplemented prairie hay (ration 1),

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and hence were restricted to this amount. Animals receiving Rations 7 and 8 were provided slightly less total feed; total organic matter of the two rations was equal. Salt was added to the rations 1 through 6 but not to 7 and 8. Rations 1 through 5 were compounded to contain 12 percent crude protein and to be as nearly equal in total digestible nutrients as practicable. It was necessary to provide small quantities of

TABLE 1. COMPOSITION OF RATIONS FED TO SHEEP PRIOR TO COLLECTION OF RUMEN CONTENTS

(Daily intake of dry matter, gm.)

	Ration Number							
Feedstuff	1	2	3	4	5	6	7	8
Alfalfa Hay					360			
Prairie Hay	600	360	360					
Sorghum Silage				360				
Corn		422	320	350	420	720	177	177
Soybean Meal			120	90	20	80		
Urea		18						
Corn Cobs							318	318
Corn Gluten Meal							141	141
Corn Syrup							49	49
Corn Oil							21	21
$CaHPO_4$							6	6
Alfalfa Ash								42
Total Amount Per Day	600	800	800	800	800	800	712	754

prairie hay to the lambs receiving the all-concentrate ration (ration 6) in order to keep them on feed. No hay was offered this lot during the three days preceding slaughter.

The animals were fed their respective rations for a period of thirty days to permit adjustment of the rumen microflora to the various diets.

Collection of samples

After this adjustment period the animals were slaughtered and the rumen contents collected for analysis. Schnambye and Phillipson (1949) had demonstrated that the concentration of volatile fatty acids in the rumen liquor of sheep was highest from 5 to 7 hours following the last feeding. These acids are formed as a result of fermentation processes in the rumen. It seemed reasonable to believe that vitamin synthesis would parallel other fermentation processes; therefore, the lambs were slaughtered approximately 6 hours following the last feeding.

Immediately following slaughter, the total contents of the rumen and reticulum were removed, weighed and large duplicate samples taken for analysis. Within one hour after collection, the samples were acidified with H_2SO_4 and autoclaved for 5 minutes to minimize further fermentation. The samples were then placed in shallow layers and dried in a forced-draft oven at 80° C. The dried material was ground in a Wiley mill.

Analytical Methods

The vitamin contents were determined by microbiological assay on acid or enzyme digests of homogenous samples of feeds and rumen contents. Hydrolysates for nicotinic acid analysis were prepared by heating 2.5 gm. of dry matter with 50 ml. of 0.1 N sulfuric acid in an autoclave for 15 minutes at 15 lb. pressure. The samples were then neutralized, diluted to a volume of 100 ml., and filtered. The filtrates were analyzed for nicotinic acid using *Lactobacillus arabinosis 17–5* essentially according to the procedure of Snell and Wright (1941). Hydrolysates for riboflavin determination were prepared by heating 2.5 gm. of dry material with 50 ml. of 0.1 N hydrochloric acid in an autoclave for 20 minutes at 15 lb. pressure. These samples were then neutralized using 0.5 N potassium hydroxide to prevent sodium chloride toxicity (MacLeod and Sneli, 1948), diluted to a volume of 100 ml., and filtered. The filtrates were analyzed for riboflavin using *Lactobacillus casei* and essentially the procedure of Snell and Strong (1939). For the determination of pantothenic acid, the digestion procedure used was that of Ives and Strong (1946) employing "Mylase P." One gm. of dry material was suspended in 50 ml. of distilled water, autoclaved for 15 minutes at 15 lb. pressure, cooled to room temperature, and 0.1 gram of "Mylase P" added. The solution was buffered with 2.5 ml. of 2.5 M sodium acetate, and the pH adjusted to 4.8 using a Beckman pH meter. The mixture was then layered with toluene, corked, and incubated for 24 hours at 50° C. At the end of the incubation period the samples were diluted to 100 ml., readjusted to a pH of 4.8, and filtered. The filtrates were then analyzed for pantothenic acid using *Lactobacillus casei* and the medium and procedure of Barton-Wright (1945).

It is recognized that the currently available methods of hydrolysis and digestion, such as were employed in this study, result in lower than the true values due to partial destruction of the vitamins during the drying process and to failure of the digestion procedure to free

all of the bound form of pantothenic acid. Values presented in table 2 should, therefore, be regarded as minimal, particularly in the case of riboflavin and pantothenic acid. Such minimum values, nevertheless, are useful in evaluating the effects of different factors on production of the vitamins by the bacteria of the rumen. The values given for nicotinic acid are the averages of at least two assays on a single hyrolysate, and those for riboflavin and pantothenic acid are averages of assays from two separate hydrolysates and digestions.

Results and Discussion

The results of this experiment are presented in table 2. Apparent synthesis of these vitamins in the rumen was computed by subtracting the vitamin content per gm. of dry ration from the vitamin content per gm. of dry rumen contents. To permit more direct comparison of the modifying effect of ration variation, all values are reduced to a common base by expressing the apparent synthesis as percentage change with respect to the ration.

Information regarding the effect of the type of roughage fed on the subsequent B vitamin complex synthesis in the rumen may be obtained by comparing the results for rations 3, 4, and 5. These rations contained corn and soybean meal and differed only in the quantities of these two concentrates and in the type of roughage. It appears that the type of roughage fed had a direct effect on the amount of B vitamin synthesis in the rumen. For example, 127 and 115 micrograms of nicotinic acid per gm. of dry rumen contents were synthesized when sorghum silage (ration 4) and alfalfa hay (ration 5) were fed, respectively. When prairie hay, which is a lower quality roughage, was fed (ration 3) only 84 micrograms of nicotinic acid were synthesized per gm. of dry rumen contents. The trend was somewhat different for riboflavin. The rumen contents obtained from lambs fed the sorghum silage ration (ration 4), the alfalfa ration (ration 5) and the prairie hay ration (ration 3) contained 10.5, 6.5, and 7.1 micrograms of synthesized riboflavin per gm. of dried material, respectively. The corresponding amounts of pantothenic acid synthesis which occurred in the rumen were 1.2, 6.1 and 8.0 micrograms per gm. of dried rumen material.

Nicotinic acid synthesis appeared to be greater when a high quality roughage such as alfalfa hay (ration 5) or sorghum silage (ration 4) was fed than when a ration which contained only concentrates was fed (ration 6). Pantothenic acid synthesis, however, was the greatest when an all-concentrate ration was fed. The amounts of B vitamins syn-

TABLE 2. RIBOFLAVIN, NICOTINC ACID AND PANTOTHENIC ACID OF RUMEN CONTENTS OF SHEEP FED DIETS OF VARYING COMPOSITION (MICROGRAMS PER GRAM OF DRY MATTER)

Ration	Ration Composition	Riboflavin			Nicotinic Acid			Pantothenic Acid		
		Ration	Rumen Contents	Change %	Ration	Rumen Contents	Change %	Ration	Rumen Contents	Change %
1	Prairie hay	3.3	6.2	88	14.5	51.4	254	10.1	8.3	—18
2	Prairie hay corn, urea	2.2	8.6	290	15.2	84.0	453	8.0	14.3	78
3	Prairie hay corn, soybean meal	2.4	9.5	296	16.4	100.4	513	9.5	17.5	84
4	Silage, corn soybean meal	3.2	13.7	327	21.1	148.6	604	15.2	16.4	8
5	Alfalfa, corn soybean meal	5.2	11.7	125	23.4	138.8	493	16.5	22.6	37
6	Corn soybean meal	1.5	9.9	560	16.9	108.4	542	7.4	30.1	306
7	Corn cob ration	1.8	6.4	256	18.6	68.2	267	5.2	16.4	215
8	Corn cob ration + alfalfa ash	1.8	8.2	356	18.6	86.9	367	5.2	18.7	260

thesized in the rumen of lambs fed the all-concentrate ration (ration 6) were nicotinic acid 91.5, riboflavin 8.4, and pantothenic acid 22.7 micrograms per gm. of dried rumen material.

B vitamin synthesis in the rumen was relatively slight when prairie hay was fed alone. The amounts synthesized were nicotinic acid 36.9, riboflavin 2.9, and pantothenic acid —2.2 micrograms per gm. of dried rumen material. The addition of corn and soybean meal to a prairie hay roughage ration (ration 1 vs. ration 3) increased the amount of synthesis of the three B vitamins markedly. These findings are in agreement with those of Lardinois *et al.* (1944) using a ration containing timothy hay as the roughage. They found that addition of readily available carbohydrate such as corn, molasses or starch and a nitrogen supplement such as urea or casein increased the synthesis of nicotinic acid, riboflavin, and pantothenic acid in the rumen.

There appeared to be slightly more rumen synthesis of the three B-vitamins when soybean meal was fed (ration 3), than when urea was fed (ration 2).

Apparently the addition of alfalfa ash to a ration containing corn cobs as the principal roughage improves the digestibility of the organic matter in the ration. It seemed desirable to study the effect of alfalfa ash on the synthesis of B-vitamins by the microorganisms present in the rumen of lambs. The results obtained in this study indicate that vitamin synthesis in the rumen was increased when alfalfa ash was added to the corn cob basal ration (rations 7 and 8). The values obtained for the cob ration (Ration 7) and the cob ration plus alfalfa ash, respectively, were as follows: nicotinic acid 49.6 and 68.3 riboflavin 4.7 and 6.5, and pantothenic acid 10.1 and 12.7 micrograms per gm. of dried rumen material.

These findings are in general agreement with those of Hunt *et al.* (1943) who found that adding readily available carbohydrates in the form of corn increased the synthesis of riboflavin in steers. In later studies Hunt *et al.* (1952), using "artificial rumen" techniques, found that the addition of starch increased the production of riboflavin, nicotinic acid, pantothenic acid and vitamin B_{12}.

That the addition of alfalfa ash increased vitamin synthesis parallels the results of *in vitro* studies by Burroughs *et al.* (1950) on the utilization of urea and the digestion of cellulose. It is also in agreement with Hunt *et al.* (1952), who found that supplementation of rumen fluids with calcium, phosphorus, and the trace minerals increased the synthesis of several vitamins of the B-complex in the artificial rumen.

Summary

Synthesis of riboflavin, nicotinic acid and pantothenic acid by the rumen microorganisms *in vivo* as affected by variations in the composition of the ration was studied, using sheep. Addition of nitrogen (either as soybean meal or urea) together with corn caused a sharp increase in the synthesis of all vitamins by animals fed a basal diet of prairie hay. Substitution of sorghum silage or alfalfa hay for prairie hay in the ration did not markedly change the absolute amounts of the vitamins present in the rumen material, although the percentage increase during the rumen fermentation was variable. Addition of alfalfa ash to a ration containing corn cobs as the principal roughage resulted in increased synthesis of all vitamins with respect to the changes occurring in the unsupplemented ration.

Literature Cited

Barton-Wright, E. C. 1945. The theory and practice of the microbiological assay of the vitamin B-complex. The Analyst 70:283.

Burroughs, W. A., P. D. Latona, P. Gerlaugh and R. M. Bethke. Mineral influences upon urea utilization and cellulose digestion by rumen microorganisms using the artificial rumen technique. JOURNAL OF ANIMAL SCIENCE 10:693.

Hunt, C. H., E. W. Burroughs, R. M. Bethke, A. F. Schalk and Paul Gerlaugh. 1943. Further studies on riboflavin and thiamine in the rumen content of cattle. II. Jour. Nutr. 25:207.

Hunt, C. H., O. G. Bentley and T. Hershberger. 1952. Factors affecting the synthesis of certain members of the vitamin B-complex in an artificial rumen. Fed. Proc. 11:233.

Ives, M. and F. M. Strong. 1946. Preparation of samples for the microbiological assay of pantothenic acid. Arch. Biochem. 9:251.

Lardinois, C. C., R. C. Mills, C. A. Elvehjem and E. B. Hart. 1944. Rumen synthesis of the vitamin B complex as influenced by ration composition. Jour. Dairy Sci. 27:579.

MacLeod, R. A. and E. E. Snell. 1948. The effect of related ions on the potassium requirement of bacteria. Jour. Biol. Chem. 176:39.

Schambye, P. and A. T. Phillipson. 1949. Volatile fatty acids in portal blood of sheep. Nature 164:1094.

Snell, E. E. and L. D. Wright. 1941. A microbiological method for the determination of nicotinic acid. Jour. Biol. Chem. 139:675.

Snell, E. E. and F. M. Strong. 1939. A microbiological assay for riboflavin. Ind. Eng. Chem., Anal. Ed. 11:346

COMPARISON OF UREA AND PROTEIN MEALS AS NITROGEN SOURCES FOR RUMEN MICRO-ORGANISMS: UREA UTILIZATION AND CELLULOSE DIGESTION

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THE utilization of urea as a partial source of protein in properly balanced rations for ruminants is a well established practice both in this country and abroad. The rumen bacteria utilize the ammonia from the hydrolysis of urea in the synthesis of their cellular protein, which in turn serves as a protein source to the host animal. In a a similar fashion the ammonia nitrogen from feed protein degradation in the rumen is fixed as bacterial protein nitrogen (McNaught and Smith, 1947). The magnitude of this conversion of dietary nitrogen to bacterial protein nitrogen has recently been reported by McDonald (1954). He demonstrated that approximately 40 percent of the zein, which contributed 94 percent of the total dietary nitrogen was utilized by the rumen microorganisms for the synthesis of their protein. Loosli *et al.* (1949) were able to maintain growth in lambs with rations in which virtually all the nitrogen was supplied by urea. They also demonstrated that the ten amino acids usually regarded as essential to growth were synthesized in the rumen of these lambs. Duncan and coworkers (1953) confirmed the synthesis of amino acids in the rumen from urea. They also showed that the mixed proteins synthesized from urea were similar to those found in the rumen of a calf on a natural ration. Burroughs *et al.* (1951) noted in their *in vitro* studies that the addition of urea to purified proteins and protein meals resulted in increased cellulose digestion and efficient urea utilization.

The objective of this study was to investigate the utilization of urea and its relation to cellulose digestion by the use of the artificial rumen technique. The specific program was (1) to compare the availability of urea nitrogen with the protein nitrogen of soybean, cottonseed, linseed, and corn gluten meals to the rumen microorganisms, (2) determine the efficiency of utilization of urea in combination with

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these protein meals, (3) determine cellulose digestion with each source of nitrogen and (4) determine the effect of urea concentration on urea utilization and cellulose digestion.

TABLE 1. COMPARISON OF UREA AND SOYBEAN MEALS AS NITROGEN SOURCES IN THE ARTIFICIAL RUMEN

	Average of 24-Hour Data				
	Ammonia Level	Urea Nitrogen Utilization		Cellulose Digestion	
Nutrient Variable	(Mg.)	(Mg.)	(%)	(Gm.)	(%)
No Nitrogen	10	..	..	2.71	30
Urea:					
(93.5 mg. N)	19	94	100	5.19	58
(187 mg. N)	61	164	88	7.26	81
(374 mg. N)	361	196	52	7.70	86
Soybean Meal No. 1:					
(93.5 mg. N)	21	..	..	4.32	48
(187 mg. N)	46	..	..	6.91	77
(374 mg. N)	222	..	..	7.37	82
1:1 Urea-Soybean Meal No. 1 Mixtures:					
(187 mg. N)	46	97	104	7.15	80
(374 mg. N)	260	115	62	7.58	85
No Nitrogen	10	..	..	2.71	30
Urea:					
(93.5 mg. N)	19	94	100	5.19	58
(187 mg. N)	93	150	80	7.53	84
(374 mg. N)	375	183	49	7.57	84
Soybean Meal No. 2:					
(93.5 mg. N)	17	..	..	4.17	47
(187 mg. N)	49	..	..	5.34	59
(374 mg. N)	231	..	..	7.26	81
1:1 Urea-Soybean Meal No. 2 Mixtures:					
(187 mg. N)	49	98	105	6.52	73
(374 mg. N)	317	102	55	7.23	81

Experimental

The technique used was essentially the same as that described by Burroughs *et al.* (1950) and Belasco (1954). Urea and the various protein meals were tested on an equivalent nitrogen basis at levels of 93.5 mg., 187 mg., and 354 mg. nitrogen, respectively, per flask per day. Also tested were 1:1 urea-protein meal mixtures, each com-

ponent contributing 93.5 mg. or 187 mg. nitrogen to the system each day. Starch was added to the flasks containing urea to compensate for the nitrogen-free extract of the protein meals, since Arias and his

TABLE 2. COMPARISON OF UREA AND CORN GLUTEN MEALS AS NITROGEN SOURCES IN THE ARTIFICIAL RUMEN

	Average of 24-Hour Data				
		Urea Nitrogen Utilization		Cellulose Digestion	
Nutrient Variable	Ammonia Level (Mg.)	(Mg.)	(%)	(Gm.)	(%)
No Nitrogen	4	..	..	2.19	25
Urea:					
(93.5 mg. N)	24	94	100	5.67	64
(187 mg.N)	96	153	82	7.00	78
(374 mg. N)	358	195	52	7.09	79
Corn Gluten Meal No. 1:					
(93.5 mg. N)	11	..	..	3.75	42
(187 mg. N)	19	..	..	5.19	58
(374 mg. N)	45	..	..	6.06	68
1:1 Urea-Corn Gluten Meal No. 1 Mixtures:					
(187 mg. N)	43	95	102	6.44	72
(374 mg. N)	165	135	72	7.42	83
No Nitrogen	6	..	..	2.70	30
Urea:					
(93.5 mg. N)	18	86	92	5.82	65
(187 mg. N)	59	172	92	7.45	83
(374 mg. N)	209	241	64	7.38	82
Corn Gluten Meal No. 2:					
(93.5 mg. N)	13	..	..	3.85	43
(187 mg. N)	20	..	..	5.33	59
(374 mg. N)	48	..	..	6.24	70
1:1 Urea-Corn Gluten Meal No. 2 Mixtures:					
(187 mg. N)	31	98	105	6.85	76
(374 mg. N)	95	174	93	7.92	88

coworkers (1951) have demonstrated that the amount of readily available carbohydrate affects the rate of nitrogen fixation and cellulose digestion.

The extent of nitrogen fixation in the systems containing urea was calculated from the diminution of urea and ammonia determined

analytically (Burroughs *et al.* 1951). Similar data could not be determined in systems with soybean meal or other protein meals since the feed protein nitrogen is not analytically distinguishable from the

TABLE 3. COMPARISON OF UREA AND LINSEED MEALS AS NITROGEN SOURCES IN THE ARTIFICIAL RUMEN

	Average of 24-Hour Data				
		Urea Nitrogen Utilization		Cellulose Digestion	
Nutrient Variable	Ammonia Level (Mg.)	(Mg.)	(%)	(Gm.)	(%)
No Nitrogen	4	..	..	1.82	21
Urea:					
(93.5 mg. N)	15	98	105	5.03	56
(187 mg. N)	66	163	87	6.59	74
(374 mg. N)	296	197	53	6.79	76
Linseed Meal No. 1:					
(93.5 mg. N)	12	..	..	4.22	47
(187 mg. N)	40	..	..	5.34	60
(374 mg. N)	187	..	..	5.60	63
1:1 Urea-Linseed Meal No. 1 Mixtures:					
(187 mg. N)	39	96	103	7.02	78
(374 mg. N)	199	123	66	7.23	81
No Nitrogen	6	..	..	2.28	26
Urea:					
(93.5 mg. N)	21	95	102	5.57	62
(187 mg. N)	83	157	84	7.46	83
(374 mg. N)	327	208	56	7.64	85
Linseed Meal No. 2:					
(93.5 mg. N)	18	..	..	5.01	56
(187 mg. N)	48	..	..	6.90	77
(374 mg. N)	171	..	..	7.38	82
1:1 Urea-Linseed Meal No. 2 Mixtures:					
(187 mg. N)	53	94	100	7.41	83
(374 mg. N)	266	104	56	7.91	88

bacterial protein nitrogen. It was necessary, therefore, to use the cellulose digestion data as an index of nitrogen utilization or availability. This index of nitrogen availability is believed valid under the conditions of these experiments, in view of the linear relationship noted

between the rate of cellulose digestion and the amount of protein or urea nitrogen added in these systems where nitrogen was the limiting factor.

TABLE 4. COMPARISON OF UREA AND COTTONSEED MEALS AS NITROGEN SOURCES IN THE ARTIFICIAL RUMEN

	Average of 24-Hour Data				
		Urea Nitrogen Utilization		Cellulose Digestion	
Nutrient Variable	Ammonia Level (Mg.)	(Mg.)	(%)	(Gm.)	(%)
No Nitrogen	7	..	..	2.31	26
Urea:					
(93.5 mg. N)	21	92	98	5.33	60
(187 mg. N)	72	161	86	7.16	80
(374 mg. N)	331	213	57	7.54	84
Cottonseed Meal No. 1:					
(93.5 mg. N)	13	..	..	4.37	49
(187 mg. N)	23	..	..	6.30	70
(374) mg. N)	61	..	..	7.30	81
1:1 Urea Cottonseed Meal No. 1 Mixtures:					
(187 mg. N)	32	100	107	6.98	78
(374 mg. N)	185	121	65	7.55	84
No Nitrogen	4	..	..	1.79	20
Urea:					
(93.5 mg. N)	18	95	102	4.85	54
(187 mg. N)	55	168	90	7.07	79
(374 mg. N)	300	224	60	7.70	86
Cottonseed Meal No. 2:					
(93.5 mg. N)	13	..	..	3.75	42
(187 mg. N)	28	..	..	5.79	65
(374 mg. N)	96	..	..	7.24	81
1:1 Urea-Cottonseed Meal No. 2 Mixtures:					
(187 mg. N)	32	99	106	6.64	74
(374 mg. N)	186	127	68	7.64	85

Results

Comparison of Urea and Protein Meals

In a preliminary experiment, better cellulose digestion was obtained with urea than with soybean meal at equivalent nitrogen levels. More detailed studies confirmed this finding. The results shown in tables

1, 2, 3 and 4 with two lots each of soybean, corn gluten, linseed and cottonseed meals indicate that urea maintained higher levels of cellulose digestion than the respective protein meals tested at comparable nitrogen levels. This was especially true at the lower nitrogen levels. At the higher nitrogen levels, where the cellulolytic response to further increases of nitrogen became less marked, the differences between

TABLE 5. THE EFFECT OF INCREASING LEVELS OF UREA IN THE ARTIFICIAL RUMEN

		Average of 24-Hour Data				
			Urea Nitrogen Utilization		Cellulose Digestion	
Urea Nitrogen Level (Mg.)	Protein Equivalent (%)	Ammonia Level (Mg.)	(Mg.)	(%)	(Gm.)	(%)
0	0	6	..	..	1.72	19
23.4	1.3	9	23	99	2.52	28
46.8	2.5	14	48	103	3.59	40
93.5	5.0	19	95	102	5.64	63
187.0	9.7	82	160	86	6.99	78
374.0	18.8	473	187	50	7.73	86
561.0	27.4	760	187	33	7.98	89
758.0	35.4	1092	195	26	8.18	91
935.0	43.0	1453	154	17	5.14	57
1122.0	50.0	1834	134	12	3.78	42
1309.0	56.7	2240	94	7	2.71	30
1516.0	63.1	2483	113	7	2.44	27

responses to urea and the protein meals, although small, were still evident.

The ammonia level increased with increasing amounts of urea and protein meals. Higher ammonia levels were noted in the flasks containing urea. The amount of urea utilized increased as the concentration of urea in the system was increased. This increase in urea utilization was paralleled by an increase in cellulose digestion. In 1:1 urea-protein meal mixtures, the percentage of urea utilized was consistently greater than the percentage utilized when urea was used alone at a similar total nitrogen level. For example, as shown in table 1, the urea in both urea-soybean meal mixtures having total nitrogen contents of 187 mg. was completely metabolized, while urea alone at the same total nitrogen levels was only 88 and 80 percent utilized in the two respective tests. This greater percentage utilization of urea in the urea-meal mixtures was noted in all the experiments. Cellulose diges-

tion rates resulting from these mixtures were comparable to those obtained with urea alone.

The Effect of Increasing Amounts of Urea in the Artificial Rumen

The effect of increasing levels of urea on the ammonia level, urea utilization, and cellulose digestion is shown in table 5. Urea levels ranged from 1.3 percent to 63 percent protein equivalent, based on the weight of nutrients used. There was a steady increase in free ammonia with increasing levels of urea. Both the urea and cellulose were metabolized in increasing amounts with increasing levels up to and including the 35 percent protein equivalent level. However, at the 43 percent protein equivalent level a sharp decrease in the amount of urea utilized and the cellulose digested was noted. Microscopic examination of bacterial smears from flasks containing these high urea levels revealed markedly reduced bacterial populations.

Discussion

It is evident that urea subject to hydrolysis by rumen bacterial urease represents a form of readily available nitrogen. On the other hand, the protein fraction of a meal represents a complex polypeptide whose nitrogen is essentially unavailable until cleavage and/or deamination occurs. Tillman and Swift (1953) speculated that the rumen microorganisms use the simpler nitrogenous materials better than preformed protein on the basis of their balance studies comparing urea, soybean meal, ammoniated cane molasses and condensed distillers molasses solubles. Sirotnak and coworkers (1953) explored the fate of various amino acids and noted that only six of the twenty-two tested were attacked by suspensions of mixed rumen organisms. Of the six amino acids, only aspartic acid was attacked at a rate comparable to that of urea.

In all the experiments reported here the cellulolytic response was greater with urea than with any of the protein meals at comparable nitrogen levels. Under the conditions of these *in vitro* experiments, the data indicate that the availability of ammonia nitrogen from either protein or nonprotein sources is an important factor in the efficient digestion of roughage. The improved roughage digestibility of rations containing urea reported by Harris and Mitchell (1941), Swift *et al.* (1947) and Briggs *et al.* (1948) possibly can be attributed to an increase of the available nitrogen of the ration in the presence of other feed proteins.

There was good correlation between the amount of urea metabolized and the amount of cellulose digested, both values showing corresponding increases up to a protein equivalent of 35 percent. At the 43 percent protein equivalent level, as shown in table 5, a sharp decrease in cellulose fermentation and urea utilization was evident. This became more marked with further increases in the urea level. It is probable that at the 43 percent protein equivalent level the hydrolysis of urea to ammonia and carbon dioxide exceeded the rate of ammonia utilization by the rumen microorganisms. Although the efficiency of urea utilization was high at the lower urea levels, it decreased with the increase of urea level. The cellulolytic response to increasing amounts of the feed proteins (tables 1, 2, 3 and 4) indicates a similar trend in nitrogen utilization by rumen microflora. According to McDonald (1949) the ammonia escaping fixation in the rumen is absorbed into the venous circulation and transported to the liver where it is converted to urea. A significant portion of this urea is returned to the rumen as a normal constituent of the saliva. This circulation of nitrogen occurring during the normal digestive process is indicative of a biological economy of ammonia arising from urea and protein degradation in the rumen.

Summary

In vitro experiments comparing urea with soybean, linseed, cottonseed and corn gluten meals indicated the superiority of urea as a nitrogen source in promoting cellulose digestion by rumen microorganisms. The utilization of urea in 1:1 mixtures with various feed proteins was very efficient, and its presence in such mixtures improved cellulose digestion above that observed with the feed proteins alone. The data indicate that the availability of nitrogen from protein or nonprotein sources is of great importance to efficient cellulose digestion.

An increase in the level of urea used as a sole source of nitrogen resulted in an increase in cellulose digestion as well as in the amount of urea utilized. A level of urea was reached above which the concentration of free ammonia apparently inhibited both urea utilization and cellulose digestion.

Literature Cited

Arias, C., W. Burroughs, P. Gerlaugh and R. M. Bethke. 1951. The influence of different amounts and sources of energy upon *in vitro* urea utilization by rumen microorganisms. JOURNAL OF ANIMAL SCIENCE 10:683.

Belasco, I. J. 1954. New nitrogen feed compounds for ruminants—a laboratory evaluation. JOURNAL OF ANIMAL SCIENCE 13:601.

Briggs, H. M., J. S. Dinning and W. D. Gallup. 1948. Feeding tests with sheep, swine and beef cattle. Okla. Agri. Exp. Sta. Misc. Pub. MP-13, 19, 28, 48.

Burroughs, W., N. A. Frank, P. Gerlaugh and R. M. Bethke. 1950. Preliminary observations upon factors influencing cellulose digestion by rumen microorganisms. Jour. Nutr. 40:9.

Burroughs, W., C. Arias, P. de Paul, P. Gerlaugh and R. M. Bethke. 1951. *In vitro* observations upon the nature of protein influences upon urea utilization by rumen microorganisms. JOURNAL OF ANIMAL SCIENCE 10:672.

Duncan, C. W., I. P. Agrawala, C. F. Huffman and R. W. Luecke. 1953. A quantitative study of rumen synthesis in the bovine on natural and purified rations. II. Amino acid content of mixed rumen proteins. Jour. Nutr. 49:41.

Harris, L. E. and H. H. Mitchell. 1941. The value of urea in the synthesis of protein in the paunch of ruminants. Jour. Nutr. 22:167, 183.

Harris, L. E., S. H. Work and L. A. Henke. 1943. The utilization of urea and soybean oil meal nitrogen by steers. JOURNAL OF ANIMAL SCIENCE 2:328.

Loosli, J. K., H. H. Williams, W. E. Thomas, F. H. Ferris and L. A. Maynard. 1949. Synthesis of amino acids in the rumen. Science 110:144.

McDonald, I. W. 1952. The role of ammonia in ruminal digestion of protein. Biochem. Jour. 51:86.

McDonald, I. W. 1954. The extent of conversion of food protein to microbial protein in the rumen of sheep. Biochem. Jour. 56:120.

McNaught, M. L. and J. A. B. Smith. 1947. Nitrogen metabolism in the rumen. Nutr. Abstr. Rev. 17:18.

Sirotnak, F. M., R. N. Doetsch, R. E. Brown and J. C. Shaw. 1953. Amino acid metabolism of bovine rumen bacteria. Jour. Dairy Sci. 36:1.

Swift, R. W., E. J. Thacker, A. Black, J. W. Bratzler and W. H. James. 1947. Digestibility of rations for ruminants as affected by proportions of nutrients. JOURNAL OF ANIMAL SCIENCE 6:432.

Tillman, A. D. and R. W. Swift. 1953. The utilization of ammoniated by-products and urea by sheep. JOURNAL OF ANIMAL SCIENCE 12:201.

COMPARISON OF UREA AND PROTEIN MEALS AS NITROGEN SOURCES FOR RUMEN MICROORGANISMS: THE PRODUCTION OF VOLATILE FATTY ACIDS

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IN recent years the importance of the formation of volatile fatty acids in the rumen for optimum nutrition of ruminants has become increasingly evident. At present, the major fatty acids such as acetic, propionic and butyric acids are considered as energy sources and key anabolic intermediates rather than waste products of digestion. Propionic acid, in particular, has received special attention as an intermediate in the biosynthesis of glucose and other carbohydrate material in the ruminant (Schultz, 1952; Pennington, 1952; Elsden *et al.*, 1948).

Because of the importance of the volatile fatty acids and their close relationship to cellulose digestion, a study was undertaken to determine the distribution of these acids under the same conditions as prevailed in a previous study comparing urea and protein meals in promoting cellulose digestion (Belasco, 1954b). The artificial rumen technique was used since it permitted a quantitative approach to this study of the fatty acids without the usual *"in vivo"* difficulties of absorption or passage of these metabolites from the rumen.

Experimental

Artificial Rumen Technique

The artificial rumen technique used in this study was the same as that described by Belasco (1954a), which basically is the technique of Burroughs *et al.* (1950). Urea and the protein meals (soybean, linseed, cottonseed and corn gluten meals) were added on an equivalent nitrogen basis of 187 mg. of nitrogen, per flask per day. As stated in a previous article (Belasco, 1954a) the input of readily available carbohydrate was balanced in all flasks, taking into account the carbohydrate fractions of the meals, in view of the influence of this nutrient on the rate of nitrogen fixation and cellulose digestion in the rumen (Arias *et al.*, 1951).

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Chromatographic Technique

The technique employed was similar to that of Linke (1952) in separating the volatile fatty acids in the rumen. This technique proved useful since it permitted separation of valeric and caproic acids from butyric acid. The apparatus was similar to that usually used in chromatographic analysis of fatty acids, employing an inert gas pressure to maintain the desired solvent flow even during the addition of eluting solvents. The chromatographic tube had an inside diameter of 20 mm. and was 40 cm. long.

Reagents

Silica gel from Mallinckrodt's silicic acid served as the partitioning agent. The silicic acid was thrice suspended in water and the unsettled portions were discarded. The coarser particles remained as components of the gel, thus reducing minimum pressure required for proper solvent flow. The moist gel was then dried for 24 hours at 100° C.

Eluting solvents were prepared in proper proportions and equilibrated with distilled water and 0.5 N H_2SO_4. The solvent ratios were 0, 1, 5 and 10 percent n-butanol in benzene. The n-butanol was freshly distilled over potassium carbonate. The solvent mixtures were passed through filter paper to remove suspended water droplets.

Cresol red indicator was made by adding 200 mg. of solid to 10.4 ml. of 0.05 N NaOH and diluting the resultant solution to 100 ml. with distilled H_2O. Standard 0.02 N NaOH (carbonate free) was used to titrate the amount of acid flowing from column; 5 N NaOH, 5 N and 0.5 N H_2SO_4 solutions were used in the preparation of samples, column and eluting solutions.

Experimental

Preparation of Sample

A 5-ml. sample of supernatant rumen liquid containing the free organic acids was placed in a 50 ml. beaker to which was added 2–3 drops of cresol red indicator. The sample was made alkaline with 5 N NaOH and evaporated to near dryness on a hot plate. Just prior to analysis, 0.5 ml. of 0.5 N H_2SO_4 was added to the dry sample containing the sodium salts of the organic acids. This was followed by the addition of 5 N H_2SO_4 dropwise until the pH was about 1 as shown by the red-orange color of the indicator. At this point 2 to 3 grams of dried silica gel were added and mixed thoroughly with the

sample to provide a free flowing powder for quantitative transfer to the top of the chromatographic column.

Column Preparation

Twenty grams of dried silicic acid was triturated in a mortar with as much 0.5 N H_2SO_4 as permissable and still maintain a powdery and free flowing mixture. Under these conditions, an additional 0.5 ml. of acid would result in a wet-appearing mixture. Approximately 75 ml. of washed benzene was added and vigorously mixed to form a slurry which was then transferred to the chromatographic tube having a cotton or glass wool plug at its base. The suspended silica gel in the tube was gently stirred with a long glass rod to remove any air bubbles. A slight positive pressure was applied with nitrogen gas, the stopcock at the bottom of the chromatographic tube being opened to permit drainage of the excess benzene from the column until its level was about 1–2 cm. above the top of the silica gel. The powdered mixture of silicic acid and volatile fatty acids to be analyzed was introduced on the top of the column through a long stemmed funnel and carefully integrated with a stirring rod into the top 1 cm. of the column.

Development of Chromatogram

Development of the column proceeded by the addition in turn of 150 ml. each of 0, 1, 5 and 10 percent n-butanol in benzene from a reservoir. By means of an inert gas pressure system, the addition of the various eluting solvents to the reservoir or from the reservoir to the column was possible without interrupting constant solvent flow. The effluent passed through an intermittent siphon (10 ml. capacity) at a uniform rate of flow of 120 ml. per hour. The individual 10 ml. fractions were collected in 100 ml. Erlenmeyer flasks containing 15–20 ml. H_2O and titrated with standard 0.02 N NaOH.

In a single trial, isolation and recovery of organic acids from a known mixture of valeric, butyric, propionic, acetic and formic acids (table 1) gave excellent results when the micro amounts involved are taken into consideration. The recovery of valeric acid was less than that of the other acids. Typical chromatograms of a mixture of volatile fatty acids in pure solution and in a sample of rumen liquid obtained by this technique are shown in figure 1.

Results

Table 2 expresses the fatty acid data on the basis of milliequivalents

TABLE 1. SEPARATION AND RECOVERY OF ORGANIC ACIDS THROUGH SILICA GEL COLUMN

Acid	Amount Added (meq.)	Amount Recovered (meq.)	Recovery (%)
Valeric	0.051	0.044	86
Butyric	0.063	0.063	100
Propionic	0.087	0.083	96
Acetic	0.139	0.142	102
Formic	0.115	0.110	96

of acid per 100 ml. rumen liquid found with the various treatments. The inoculum represents a sample of the rumen liquid from a fistulated steer which had been without feed for approximately 16 hours prior to sampling. It served as the original inoculum on the first day of

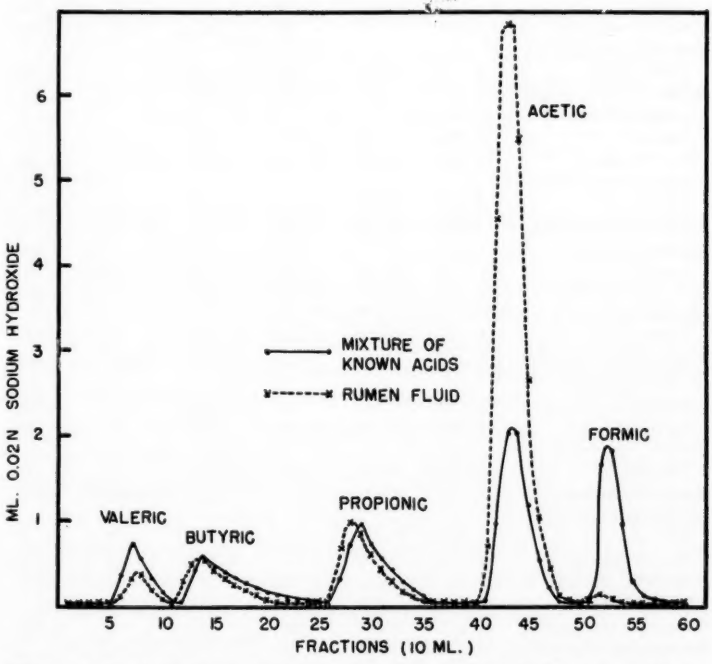

Figure 1. Typical chromatograms of a mixture of volatile fatty acids in pure solution and in a sample of rumen liquid.

TABLE 2. VOLATILE FATTY ACID FORMATION IN THE ARTIFICIAL RUMEN

Items	Milliequivalents of Acid per 100 ml. Rumen Liquid					
	Valeric	Butyric	Propionic	Acetic	Formic	Total
Rumen liquid inoculum	0.192	0.545	0.802	4.112	0	5.651
No nitrogen (neg. control)	0	0.217	1.235	2.766	0	4.218
Urea (187 mg. N)	0.053	0.509	5.795	6.601	0	12.958
Soybean Meal No. 1 (187 mg. N)	0.198	0.695	5.290	7.190	0	13.373
Rumen liquid inoculum	0.192	0.545	0.802	4.112	0	5.651
No nitrogen (neg. control)	0	0.217	1.235	2.766	0	4.218
Urea (187 mg. N)	0	0.479	5.730	6.403	0	12.612
Soybean Meal No. 2 (187 mg. N)	0.133	0.559	4.693	6.677	0	12.062
Rumen liquid inoculum	0.221	0.475	1.068	4.714	0.211	6.689
No nitrogen (neg. control)	0.065	0.451	1.478	3.122	0	5.116
Urea (187 mg. N)	0.145	0.731	4.706	6.602	0	12.181
Corn gluten meal No. 1 (187 mg. N)	0.256	0.857	4.308	6.281	0	11.702
Rumen liquid inoculum	0.195	0.851	0.708	3.764	0	5.590
No nitrogen (neg. control)	0.065	0.560	2.034	3.155	0	5.814
Urea (187 mg. N)	0.142	0.650	6.139	7.284	0.073	14.288
Corn gluten meal No. 2 (187 mg. N)	0.292	0.804	4.734	6.338	0.106	12.274
Rumen liquid inoculum	0.166	0.530	0.664	2.952	0.231	4.543
No nitrogen (neg. control)	0	0.540	1.453	2.347	0	4.340
Urea (187 mg. N)	0.122	0.581	5.790	6.569	0	13.062
Linseed meal No. 1 (187 mg. N)	0.203	0.800	5.144	7.271	0	13.418
Rumen liquid inoculum	0.207	0.771	0.944	4.798	0.068	6.838
No nitrogen (neg. control)	0	0.407	1.136	2.858	0	4.401
Urea (187 mg. N)	0.068	0.574	5.601	7.205	0	13.448
Linseed meal No. 2 (187 mg. N)	0.258	0.863	5.008	6.988	0	13.117
Rumen liquid inoculum	0.168	0.814	1.119	4.758	0.118	6.977
No nitrogen (neg. control)	0.081	0.633	1.993	2.866	0.097	5.670
Urea (187 mg. N)	0.089	0.560	5.745	6.723	0.171	13.288
Cottonseed meal No. 1 (187 mg. N)	0.288	0.885	5.164	7.381	0.508	14.226
Rumen liquid inoculum	0.173	0.494	0.806	3.523	0	4.996
No nitrogen (neg. control)	0	0.293	1.148	2.223	0	3.664
Urea (187 mg. N)	0	0.617	5.302	6.845	0	12.764
Cottonseed meal No. 2 (187 mg. N)	0.150	0.682	3.804	6.695	0	11.331

fermentation. Chromatographic data were obtained on samples at the end of the third day of fermentation from the flasks receiving urea and the protein meal, and from the negative control flask which received no nitrogen supplements.

Examination of the data reveals that urea, as the nitrogen substrate, increased propionic acid and decreased butyric acid production when compared to the various protein meals. This difference was consistent in all the experiments. Wherever the data permitted comparison, valeric acid production was also consistently higher in the flasks containing the protein meals. In general, the results indicate that acetic acid production was unaffected by type of nitrogen source used. Similar relationships are evident in the molar ratios of the three major organic acids (table 3). All the ratios, propionic/butyric, propionic/acetic and acetic/butyric, were consistently greater with urea than with the protein meals.

As summarized in table 4, it is evident that propionic acid production was from 9 to 39 percent greater in the flasks containing urea than in those containing the protein meals. Similarly, butyric acid production was from 10 to 37 percent less in flasks containing urea.

Discussion

It is obvious from the results that urea as the sole nitrogen substrate led to more propionic acid and less butyric and valeric acids than the equivalent amounts of protein feed meals. The higher levels of butyric and valeric acids found with the meals was probably due, in part, to deaminated amino acid fragments of the meal proteins. El-Shazly (1952) found that some of the higher volatile fatty acids formed in the rumen originate mainly from the breakdown of protein. Since the structure of urea did not contribute to any of the volatile fatty acids, it would be reasonable to assume that the increase of propionic acid noted may result from more efficient cellulose digestion in the presence of urea (Belasco, 1954b). In other words, the hydrolysis of cellulose in the presence of urea in the rumen may normally give rise to proportionately greater amounts of propionic acid than the other acids.

Propionic acid is utilized by most ruminant tissues in the biosynthesis of glucose, glycogen and other carbohydrate materials (Pennington, 1952) and is therefore considered glycogenic. It is probable that propionate is the major carbohydrate source for these animals, since

little of the available dietary carbohydrates escapes fermentation. The data presented by Phillipson (1947) support this view.

Acetic acid and especially butyric acid, although anabolic intermediates, are ketogenic. Jarrett and Potter (1950) administered acetate and propionate to sheep and found that acetate increased while pro-

TABLE 3. MOLAR RATIOS OF VOLATILE FATTY ACIDS IN THE ARTIFICIAL RUMEN

Items	Acid Ratios		
	Propionic / Butyric	Propionic / Acetic	Acetic / Butyric
Rumen liquid inoculum	1.47	0.20	7.54
No nitrogen (neg. control)	5.69	0.45	12.75
Urea (187 mg. N)	11.39	0.88	12.97
Soybean meal No. 1 (187 mg. N)	7.61	0.74	10.35
Rumen liquid inoculum	1.47	0.20	7.54
No nitrogen (neg. control)	5.69	0.45	12.75
Urea (187 mg. N)	11.96	0.89	13.37
Soybean meal No. 2 (187 mg. N)	8.40	0.70	11.94
Rumen liquid inoculum	2.25	0.23	9.92
No nitrogen (neg. control)	3.28	0.47	6.92
Urea (187 mg. N)	6.44	0.71	9.03
Corn gluten meal No. 1 (187 mg. N)	5.03	0.69	7.33
Rumen liquid inoculum	0.92	0.21	4.42
No nitrogen (neg. control)	3.62	0.65	5.63
Urea (187 mg. N)	9.44	0.84	11.21
Corn gluten meal No. 2 (187 mg. N)	5.89	0.75	7.88
Rumen liquid inoculum	1.25	0.22	5.57
No nitrogen (neg. control)	2.69	0.62	4.35
Urea (187 mg. N)	9.97	0.88	11.31
Linseed meal No. 1 (187 mg. N)	6.43	0.71	9.09
Rumen liquid inoculum	1.29	0.21	6.22
No nitrogen (neg. control)	2.79	0.40	7.02
Urea (187 mg. N)	9.76	0.78	12.55
Linseed meal No. 2 (187 mg. N)	5.80	0.72	8.10
Rumen liquid inoculum	1.37	0.24	5.85
No nitrogen (neg. control)	3.15	0.70	4.52
Urea (187 mg. N)	10.26	0.85	12.01
Cottonseed meal No. 1 (187 mg. N)	5.84	0.70	8.34
Rumen liquid inoculum	1.63	0.23	7.13
No nitrogen (neg. control)	3.92	0.52	7.59
Urea (187 mg. N)	8.59	0.77	11.09
Cottonseed meal No. 2 (187 mg. N)	5.58	0.57	9.82

pionate decreased the level of ketone bodies in the blood. Simultaneous administration of acetate and propionate produced little rise in ketone bodies, suggesting that the oxidation of acetate is closely associated

TABLE 4. COMPARISON OF UREA AND PROTEIN MEALS IN THE FORMATION OF THE MAJOR FATTY ACIDS IN THE ARTIFICIAL RUMEN

Items	Nitrogen Equivalent Added per Day	Milliequivalents of Acid per 100 ml. Rumen Liquid					
		Butyric	Diff.	Propionic	Diff.	Acetic	Diff.
	mg.		%		%		%
Urea	187	0.509	−27	5.795	+10	6.601	−8
Soybean meal No. 1	187	0.695		5.290		7.190	
Urea	187	0.479	−14	5.730	+22	6.403	−4
Soybean meal No. 2	187	0.559		4.693		6.677	
Urea	187	0.731	−15	4.706	+9	6.602	+5
Corn gluten meal No. 1	187	0.857		4.308		6.281	
Urea	187	0.650	−19	6.139	+30	7.284	+15
Corn gluten meal No. 2	187	0.804		4.734		6.338	
Urea	187	0.581	−27	5.790	+13	6.569	−10
Linseed meal No. 1	187	0.800		5.144		7.271	
Urea	187	0.574	−34	5.601	+12	7.205	+3
Linseed meal No. 2	187	0.863		5.008		6.988	
Urea	187	0.560	−37	5.745	+11	6.723	−9
Cottonseed meal No. 1	187	0.885		5.164		7.381	
Urea	187	0.617	−10	5.302	+39	6.845	+2
Cottonseed meal No. 2	187	0.682		3.804		6.695	

with the metabolism of propionate. Schultz *et al.* (1949) reported that the administration of butyric and some of the higher acids to goats proved ketogenic. Recent studies by Pennington (1952) conclusively demonstrated that the epithelium of the rumen, reticulum, and omasum, as well as liver tissue, are sites for the conversion of butyric and acetic acids to ketone bodies. Propionic acid was unaffected. He postulates that the propionates in the intact animal facilitate the complete

oxidation of the other fatty acids and thus prevent the excessive formation of ketone bodies. The role of a dietary source of propionate has been further elaborated by Schultz (1952).

El-Shazly (1952) speculates that high-protein diets may be responsible for abnormally high levels of blood ketone bodies. In agreement with this speculation, the findings presented here indicate that urea as a partial protein replacement in such rations could alleviate such conditions by supporting the production of more propionic acid and less of the ketogenic butyric and valeric acids.

Summary

Urea as the sole nitrogen source in the artificial rumen promoted the formation of higher levels of propionic acid and lower levels of butyric and valeric acids than did nitrogen equivalent amounts of high protein feeds such as soybean, linseed, cottonseed and corn gluten meals. The acetic acid level and the total quantity of volatile fatty acids were unaffected by the type of nitrogen substrate. It is postulated that high propionic acid production is the result of high celluloytic activity in the rumen in the presence of a highly readily available source of nitrogen such as urea.

Literature Cited

Arias, C., W. Burroughs, P. Gerlaugh and R. M. Bethke. 1951. The influence of different amounts and sources of energy upon *in vitro* urea utilization by rumen microorganisms. JOURNAL OF ANIMAL SCIENCE 10:683.

Belasco, I. J. 1954a. New nitrogen feed compounds for ruminants—a laboratory evaluation. JOURNAL OF ANIMAL SCIENCE 13:601.

Belasco, I. J. 1954b. Comparison of urea with feed protein meals by the artificial rumen technique: urea utilization and cellulose digestion. JOURNAL OF ANIMAL SCIENCE. 13:739.

Burroughs, W., N. A. Frank, P. Gerlaugh and R. M. Bethke. 1950. Preliminary observations upon factors influencing cellulose digestion by rumen microorganisms. Jour. Nutr. 40:9.

Elsden, S. R. and A. T. Phillipson. 1938. Ruminant digestion. Ann. Rev. Biochem. 17:705.

El-Shazly, K. 1952. Degradation of protein in the rumen of the sheep. 1. Some volatile fatty acids, including branched chain isomers found *in vivo*. 2. The action of rumen microorganisms on amnio acids. Biochem. Jour. 51:640, 647.

Gray, F. V. and A. F. Pilgrim. 1952. Origins of the volatile fatty acids in the rumen. Nature 170:375.

Jarrett, I. G. and B. J. Potter. 1950. Metabolism of acetate and propionate in the ruminant. Nature 166:515.

Linke, E. G. 1952. Chromatographic separation of the volatile fatty acids produced in the rumen. M. S. Thesis, Ohio State University.

Pennington, R. J. 1952. The metabolism of short-chain fatty acids in the sheep. 1. Fatty acid utilization and ketone body production by rumen epithelium and other tissues. Biochem. Jour. 51:251.

Phillipson, A. T. 1947. Fermentation in the alimentary tract and the metabolism of the derived fatty acids. Nutr. Abstr. & Rev. 17:12.

Schultz, L. H., V. R. Smith and H. A. Lardy. 1949. The effect of the administration of various fatty acids on the blood ketone levels of ruminants. Jour. Dairy Sci. 32:817.

Schultz, L. H. 1952. Recent studies on ketosis in dairy cattle. Proc. 1952 Cornell Nutrition Conference, 79–86.

THE EFFECT OF ADDED FAT ON THE DIGESTION OF CELLULOSE AND PROTEIN BY OVINE RUMEN MICROORGANISMS [1]

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THE annual domestic surplus of tallow and grease has been estimated to be in excess of 700 million pounds, and a price outlook has been forecast which would place these materials in competition with other sources of energy in animal feeds (Ewell, 1953). Few experiments have been conducted to determine the effect of fat on rumen functions. Swift *et al.* (1948) failed to establish the optimum level of fat to use in ruminant feeds. Wiley *et al.* (1952) reported that the addition of 5 percent of cottonseed oil to practical fattening rations for cattle reduced the feed intake but increased the feed efficiency. The use of a corn oil or tallow pellet for beef cattle fattening rations has been reported by Matsushima *et al.* (1953). Allen and Fitch (1941) replaced 3.4 lb. of a starch-sucrose mixture with 1.5 lb. of various fats in dairy cattle rations. Corn oil and soybean oil depressed milk fat production, but all other added fats tested stimulated milk fat production.

This paper reports experiments designed to determine the effect of corn oil and lard on the digestion of cellulose by ovine rumen microorganisms *in vitro* and on the digestion of cellulose and crude protein *in vivo*.

Experimental

Four trials were run with the artificial rumen as described by Brooks (1954). Each artificial rumen contained 500 mg. filter paper, 40 mg. casein, 10 mg. urea, and 10 mg. ammonium carbonate. Each test rumen was paired with a control rumen containing no added fat. Corn oil (10 to 170 mg.) was dissolved in ether, mixed with the dry ingredients of the basal "ration" and the ether removed by warming on a water

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bath. The liquid phase contained 10 ml. artificial saliva (McDougal, 1948), 5 ml. distilled water and 10 ml. of rumen liquor used for inoculation. The rumen liquor used for inoculation was drawn from a fistulated sheep maintained on a high-cellulose ration. The rumen liquor was shaken under carbon dioxide and strained through cheese cloth. The tubes were incubated at 39° C. with slow agitation for 40 hours. After incubation the pH of each tube was determined, the bacteria present were compared to those initially present and tests were made to determine the composition of the end products of digestion. The observations made on the artificial rumen and the similarity between the results obtained *in vivo* and *in vitro* indicated that the conditions maintained in the artificial rumen were similar to those in the rumen of the animal. Cellulose determinations were made by the Viles (1948) anthrone method as described by Huhtanen *et al.* (1954).

Three digestion trials were conducted by the chromium oxide indicator method with 20 crossbred yearling wethers in each trial to determine the effect of adding corn oil and lard to a basal ration of 908 gm. cottonseed hulls, 94 gm. casein, 6 gm. Cr_2O_3 and 2500 I. U. vitamin A per day. The value of 18 gm. of alfalfa ash added to the rations containing corn oil or lard was also determined. The ash was prepared from third-cutting leafy, green, U. S. no. 1 alfalfa. Preliminary periods lasted 14 days and collection periods 4 days. Two 2-hour "grab samples" of feces were obtained daily by clipping a plastic freezer bag to the wethers. The feces were stored at —20° C. until the end of the 4-day collection period. The pooled collections were removed from the freezer, thawed, mixed, dried in a forced draft oven, ground, and sampled. The dried sample was stored in an air-tight container at room temperature until analyzed.

Feces and feeds were analyzed for cellulose by the Crampton and Maynard (1938) method, for Cr_2O_3 by the method of Gehrke *et al.* (1950) modified according to Bolin (1952), and for crude protein by the A.O.A.C. method. At the conclusion of trials II and III, samples of the rumen contents were drawn by stomach tube. The bacterial population was estimated by the method of Moir (1951) and volatile fatty acids (VFA) were determined at the end of trial III as described by Pfander and Phillipson (1953).

The chromic oxide was premixed with the casein and the weighed mixture was stirred into the hulls at feeding time. The mixture of chromic oxide and casein adhered to the cottonseed hulls. (Coefficients

of digestibility of organic matter, cellulose and protein obtained with chromic oxide as a reference substance do not differ significantly from the total collection method if the basal ration is carefully mixed; Crampton and Lloyd, 1951; Brooks, 1953).

Sheep had free access to salt and a mineral mixture made up of equal parts salt, ground limestone and dicalcium phosphate. Each 100 lb. of the mineral mixture contained 2 oz. cobalt sulphate.

During trial I all sheep were on the basal ration. In trials II and III there were four sheep on each ration. In trial II the following supplements were added to the basal ration: Lot 1, no addition; Lot 2, 32 gm. corn oil; Lot 3, 64 gm. corn oil; Lot 4, 32 gm. corn oil, 18 gm. alfalfa ash; Lot 5, 64 gm. corn oil, 18 gm. alfalfa ash. Corn oil was heated to 160° F. and mixed thoroughly with the hulls. In trial III lard was fed in place of corn oil. The lard contained 0.002 percent butylated hydroxy anisole and 0.001 percent propyl gallate and was added to the hulls at 160° F.

No sheep receiving the basal ration refused feed, but there were some refusals when fat was added to the ration. Following a refusal the amount of all components of the ration was reduced (the proportion of each ingredient was kept constant) until the sheep consumed all of the feed offered.

Results

The addition of corn oil significantly decreased cellulose digestion in the artificial rumen (table 1). The decrease ranged from 40 to 94 percent when 10 to 170 mg. of corn oil were added. The fat, when present in large amounts, did not emulsify and formed a layer in the artificial rumen.

Digestion trial I was a uniformity test. The mean coefficient of digestibility of cellulose was 40.96 $\pm$ 3.53, and the mean coefficient of protein digestion was 37.44 $\pm$ 3.31. There were no significant differences between lots, nor did the performance of the sheep in lot 1 in trials II and III differ significantly from their performance in trial I.

Comparisons were made between lots within trial rather than between the performances of animals in trials II and III with their performance in trial I.

Tables 2 and 3 summarize the coefficients of digestibility of cellulose and protein obtained in the digestion trials and give the number of bacteria and concentration of volatile fatty acid present in the rumen contents at the end of the trials.

TABLE 1. THE EFFECT OF ADDED CORN OIL ON CELLULOSE DIGESTION BY OVINE RUMEN ORGANISMS IN THE ARTIFICIAL RUMEN

Corn Oil (Mg.)	Trial	No. of Pairs of Rumina	Percent Cellulose Digestion		
			Basal	Basal + Corn Oil	Difference (%)
10	1	5	36.2	21.6	−40.3**
20	2	10	31.8	19.3	−39.3**
40	3	20	41.7	21.3	−48.9**
60	4	5	27.4	19.4	−29.2**
80	2	10	31.8	16.0	−49.7**
160	1	5	36.2	2.2	−93.9**
170	3	5	56.0	3.2	−94.3**

** Difference in cellulose digestion (experimental vs. basal) highly significant (P less than 0.01).

All sheep lost weight during trial II, but the weight loss was greater in sheep receiving the corn oil than in sheep fed the basal ration. The coefficients of digestibility of cellulose and crude protein were reduced 52 and 17 percent, respectively, by the addition of 32 gm. of corn oil. Two sheep developed anorexia and consumed only 87 percent as much feed as the sheep in the basal lot during the last part of the feeding period.

When 64 gm. of corn oil was fed daily, the sheep consumed only 70 percent as much feed as the basal lot and they scoured and appeared listless. The rumen contents were white and turbid, and had an abnormal

TABLE 2. THE EFFECT OF ADDED CORN OIL WITH AND WITHOUT ALFALFA ASH ON THE DIGESTION OF CELLULOSE AND CRUDE PROTEIN AND THE NUMBERS OF BACTERIA IN THE RUMEN OF THE SHEEP

Trial II

Additions to Basal Ration	Cellulose Digestion (%)	Crude Protein Digestion (%)	No. Bacteria per Gram Rumen Contents
None	41.9	37.5	21×10^9
Corn oil, 32 gm.	20.0*	30.9	19×10^9
Corn oil, 64 gm.	12.3**	23.8	20×10^9
Corn oil, 32 gm. plus Alfalfa ash, 18 gm.	40.0	34.8	.....[1]
Corn oil, 64 gm. plus Alfalfa ash, 18 gm.	21.0*	28.8	.....[1]

* Decrease in digestion was significant (P less than .05).
** Decrease in digestion was highly significant (P less than .01).
[1] No count available.

putrid odor. *Aerobacter aerogenes* was isolated from the rumen contents. The coefficients of digestibility of cellulose and crude protein were reduced 70 and 36 percent, respectively.

Sheep receiving 32 gm. of lard per day digested 33 percent less cellulose than those on the basal ration. There was no apparent loss of appetite and there was little change in the digestibility of crude protein.

TABLE 3. THE EFFECT OF ADDED LARD WITH AND WITHOUT ALFALFA ASH ON CELLULOSE AND PROTEIN DIGESTION, VOLATILE FATTY ACID CONCENTRATION AND NUMBERS OF BACTERIA IN THE RUMEN OF THE SHEEP

Trial III

Additions to Basal Ration	Cellulose Digestion (%)	Protein Digestion (%)	V.F.A. of Rumen Content (Mg. %)	No. of Bacteria per Gram Rumen Contents
None	39.9	35.8	4.1	21×10^9
Lard, 32 gm.	27.8	35.3	3.2	20×10^9
Lard, 64 gm.	19.5	35.5	2.4	19×10^9
Lard, 32 gm. plus Alfalfa ash, 18 gm.	33.9	35.2	4.3	25×10^9
Lard, 64 gm. plus Alfalfa ash, 18 gm.	34.4	36.8	4.1	22×10^9

The volatile fatty acid level in the rumen contents was lower than in sheep receiving the basal ration.

Sheep receiving 64 gm. of lard did not scour or appear listless; their feed intake was reduced 6 percent. The coefficient of digestibility of cellulose was 53 percent lower than in sheep on the basal ration, and the coefficient of digestibility of crude protein was decreased 33 percent. The rumen contents contained 2.4 mg. percent V.F.A. as compared to 4.1 mg. percent in sheep on the basal ration.

Sheep receiving fat were similar to those on the basal ration in the number of bacteria present in the rumen. There did appear to be a qualitative shift in bacteria as indicated by an increase in the number of medium-sized cocci and a decrease in the numbers of small rods in the fat-fed animals. Fat did not cause severe digestive upsets when alfalfa ash was present in the ration.

Discussion

The severe depression of cellulolytic activity when fat was added to the ration indicates that large amounts of fat should not be added to high-fiber rations if maximum cellulose digestion is desired.

The protective effect of alfalfa ash suggests the need for further study on factors determining cellulose breakdown. Part of the effect of alfalfa ash may be due to its buffering capacity or to its ability to help to emulsify the added fat and thereby prevent it from coating the cellulose fiber.

Corn oil caused a greater reduction in cellulose digestion than did lard. Allen and Fitch (1941) stressed the depressing effect of 1.5 lb. corn oil in dairy rations, and Matsushima *et al.* (1953) reported that steers fed corn oil pellets made slower gains than similar steers fed tallow pellets. The addition of corn oil to some ruminant rations does not seem to be desirable. Many practical rations, however, contain as much total fat as the rations used in these studies. Added fat apparently depresses cellulose utilization more than the fat originally present in the feed—especially when alfalfa ash is not present.

Summary

The addition of corn oil (10–170 mg.) to 1 gram of dry matter containing 50 percent cellulose in the artificial rumen inoculated with ovine rumen organisms significantly reduced cellulose digestion (40–94%).

Thirty-two or 64 grams of corn oil added to a basal cottonseed hulls and casein ration significantly reduced cellulose digestion and lowered protein digestion in sheep. Rumen ingesta had a putrid odor, a turbid color and a lowered volatile fatty acid content. The total number of bacteria present was not reduced, but there was a decrease in the number of small rods and an increase in small cocci when fat was present. *Aerobacter aerogenes* was isolated from the rumen contents of sheep fed 64 grams of corn oil.

Thirty-two or 64 grams of lard added to the basal ration also decreased cellulose digestion.

The depressing effects of added corn oil or lard were partially overcome by addition of alfalfa ash.

Literature Cited

Allen, N. N. and J. B. Fitch. 1941. The influence of sustained high fat intake upon milk fat production. Jour. Dairy Sci. 24:516.

Bolin, D. W. and R. P. King. 1952. A simplified method for determination of Cr_2O_3 when used as index. Science 116:634.

Brooks, C. C. 1953. Unpublished.

Brooks, C. C. 1954. Ph.D. Thesis. University of Missouri.

Crampton, E. W. and L. A. Maynard. 1938. The relation of cellulose and lignin content to the nutritive value of animal feeds. Jour. Nutr. 15:383.

Crampton, E. W. and L. E. Lloyd. 1951. Studies with sheep on the use of chromic oxide as an index of digestibility of ruminant rations. Jour. Nutr. 45:319.

Ewell, Raymond H. 1953. The outlook for inedible fats and oils. Jour. of Agr. and Food Chem. 1:552.

Gehrke, C. W., D. T. Mayer, E. E. Pickett and C. V. Runyon. 1950. The quantitative determination of chromic oxide in feeds and feces. U. of Mo. Res. Bul. 469.

Huhtanen, C. N., R. K. Saunders and L. S. Gall. 1954. Fiber digestion using the miniature and artificial rumen. Jour. Dairy Sci. 37:328.

Matsushima, J. and T. W. Dowe. 1953. Feed Age 3:34.

McDougal, E. I. 1948. Studies on ruminant saliva. 1. The composition and output of sheep's saliva. Biochem. Jour. 43:99.

Moir, R. J. and V. J. Williams. 1951. Ruminal flora studies in the sheep. Aust. Jour. Sci. Res. B. 3:381

Pfander, W. H. and A. T. Phillipson. 1953. The rates of absorption of acetic, propionic and n-butyric acids. Jour. Physiol. 122:1.

Swift, R. W., J. W. Bratzler, W. H. James, A. D. Tellman and D. C. Meek. 1948. The effect of dietary fat on utilization of the energy and protein of rations by sheep. JOURNAL OF ANIMAL SCIENCE 7:475.

Viles, F. J. J. and L. Silverman. 1948. The determination of starch and cellulose with anthrone. Anal. Chem. 21:950.

Wiley, N. B., J. K. Riggs, R. W. Colby, O. D. Butler and R. Reiser. 1952. The influence of fat and energy in the ration upon feedlot performance and carcass composition of fattening steers. JOURNAL OF ANIMAL SCIENCE 11:705.

SEQUENCE AND GROWTH OF PRIMARY AND SECONDARY FIBER FOLLICLES IN KARAKUL SHEEP

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THE opinion is frequently expressed that new follicles must be produced in the skin of fur- and wool-bearing animals throughout life. While biologists may doubt the validity of this view because of their realization that once a tissue is differentiated it is usually incapable of producing new organs, no final answer has thus far been offered. In this study, observations have been made on consecutive stages from foetuses 45 days old to adults of old age.

Karakul sheep seemed particularly useful animals for investigation as their fiber products are both fur and wool. Karakul lambs 1 to 5 days old produce excellent pelts popularly known as Persian lamb, while Karakul sheep are a source of carpet wool.

The object of the present studies was to answer the following questions: (1) Is there a chronological sequence of development of primary and secondary follicles? (2) Are new follicles being initiated throughout life? (3) Is there a relationship between the depth of follicular penetration in the skin with skin thickness and age? (4) Is there a difference in the shape and orientation of the follicles in regions producing the most desirable type of Karakul curl, for example, the dorsum, as compared to the ventral region showing poorer or hardly any curls?

Material

The skins used in these observations were collected from the flock of Karakul sheep at the Agricultural Research Center, Beltsville, Maryland, during the years 1949 to 1951.

Twenty-one foetuses ranging from 45 to 147 days of uterine life, 21 lambs ranging from 1 day to 7 months, and 10 sheep 1 to 8 years old, furnished the samples. Gestation in Karakul sheep is about 151 days. Of the total of these 52 animals, 10 were pure Karakuls, 40 were high-

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grade Karakuls (87.5 to 99.6 percent Karakul) and only 2 foetuses carried no more than 75 percent Karakul heredity.

Methods

Rectangular pieces of skin about 1″ × 2″ were cut out from mid-dorsum and mid-venter immediately upon the death of the animals and after the fibers had been sheared off. Rectangles of certain fetal jaws and head skins, and occasionally of other body regions of postnatal animals were also included. Smaller portions of skin were cut out of young foetuses. The fresh samples were flattened out on file cards, (the fascia acting as a form of adhesive medium), and immersed in Bouin's killing and fixing fluid. Upon fixation the samples were examined under a binocular microscope and cut in appropriate pieces so as to assure suitable orientation of the fibers. They were then dehydrated and embedded in tissue-mat, after which they were cut on the microtome at 8 microns or thicker as the case might require. The resulting sections were next affixed to slides, stained, and mounted for microscopic examination.

Microscopic measurements

A projector adjusted to a magnification of 500 X was used for measuring the thickness of the skin and follicular penetration. The epidermis was measured by means of an ocular micrometer under a magnification of 200 X.

Photography

The photomicrographs were taken at a magnification of 15 X for the vertical sections, and a magnification of 50 X for the parallel sections of the skin.

Measurement of body surface.

The surface of the body was calculated according to a formula proposed by Ritzman and Colovos (1930). The surface in square meters can be expressed thus $S = KW^r$, where K and r are constants and W the weight in kilograms of the sheep. As K and r change, depending on the age of the animals, the formula used would appear as follows:

for lambs $S = .139\,W^{.501}$
for yearlings $S = .147\,W^{.520}$
for adults $S = .126\,W^{.556}$

Observations and Discussion

That the epidermis of the skin of Karakul sheep is a relatively thin layer throughout life was confirmed by observations and measurements. The epidermis is just a few cells in thickness, this finding corresponding to descriptions by Herre and Rabes (1937), and Leshchinskaia (1952) of post-natal animals.

The germinal layer, that is, the proliferating, non-keratinized portion of the epidermis which is responsible for the formation of all the fiber follicles, is only 15 to 20 microns thick in a 70-day foetus. At that stage the body of the Karakul foetus is already covered with the anlagen of the primary follicles, and the above epidermal measurements remain fairly constant through pre-natal life.

In post-natal stages the germinal layer of the epidermis measures from 16 to 25 microns, and in an exceptionally heavy region it may reach up to 30 microns. In any case, even in unusually thick regions, the difference is no more than an extra layer of epidermal cells.

Age and sex seem to play no part so far as the thickness of the germinal layer is concerned. The layer tends to be somewhat thinner on the ventral than on the dorsal side in lambs a few months old and in mature sheep, but this does not hold true for foetal stages, including the all-important period during which the anlagen of the primary and secondary follicles are being initiated. Margolena and Dolnick (1952) noted that frequency of mitotic figures in the germinal layer decreases spectacularly towards the end of the above periods, that is, in foetuses 90 to 100 days old, and does not return again to that high level.

The Follicles

The follicles of Karakul sheep develop as typical mammalian hair follicles, that is, they begin as groups of cells set aside by the epidermis. These cells arrange themselves about a visibly undifferentiated center, thus forming the anlage or follicular bud. Three to four and a half weeks later, or in foetuses 90 to 102 days old, the bud has already developed into a partially formed follicle with growing accessory structures. These are the sebaceous and the sudoriferous glands, and the hair muscle (arrector pili), the latter is produced independently from the connective tissue cells. In another week, that is in foetuses 3½ months and up, one may notice within the follicles upward growing cones which are the future fibers that begin to show traces of keratinization.

The first anlagen are present on the crown of the head and about the jaws in approximately 60 to 65-day foetuses, and develop over the whole body towards 70 days of uterine life. Other authors have found corresponding follicular developments in sheep kept under different climatic conditions. Thus Duerden (1924) found in South Africa that Merino sheep foetuses show follicular anlagen at about 2½ months; Herre and Wigger (1939) in Germany refer to a Karakul foetus 75 days old; and Nikolskii (1932) reports for two regions in the U.S.S.R.—70 to 75 days as the initial periods of follicular formation in Karakul sheep.

The anlagen of the first complete follicular population develop in Karakul foetuses at approximately 70 days which is 3 weeks ahead of the following generation and is referred to as primary. The difference between the primary and secondary generations is, however, not just a chronological one as elucidated below.

Primary and secondary follicles. As proposed by Carter (1939) for wool sheep, each primary follicle carries a sebaceous gland, a sudoriferous gland, and a hair muscle, while the secondary follicles lack the sudoriferous gland and usually the other structures as well.

Carter's classification is quite applicable for the differentiation of follicles of Karakul sheep, if the criterion depends on the presence or absence of the sudoriferous gland. While the sudoriferous gland, or more exactly its canal, is quite persistent in retaining its original position, the location of sebaceous glands and hair muscles in older skins may be obscured by excessive lobulation of the former and displacement of the latter, all of which may contribute to mistaken identification of the given follicle.

From the time the foetus is about 3½ months old, one can distinguish the primary from the secondary follicles by size (figure 5, no. 1) as well as by shape (figure 5, nos. 2, 3, and 4). Note that the lumen in the canal of the sudoriferous gland is helpful in recognizing the gland both in longitudinal as well as in cross sections (figure 5, nos. 2, 3, and 4).

In studying parallel sections of foetal skins 3½ to 4 months old, one finds that the secondary generation of follicles usually develops from anlagen initiated in the epidermis opposite the sudoriferous gland of the primary follicle. This is verified in vertical sections of foetuses about 90 to 102 days old; the anlagen are situated in the acute corners formed by the primary follicle. The accessory structures of the primary follicle develop in the obtuse corners (figure 5a).

By the time the anlagen of the secondary follicles emerge, the primary follicles have usually already attained a considerable degree of

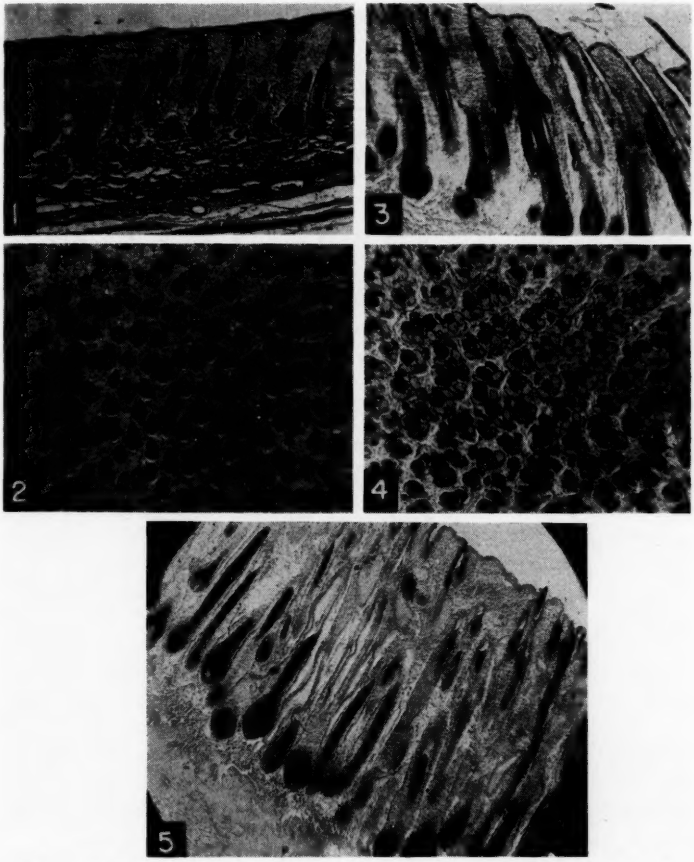

Figure 5. Skin sections of mid-dorsal area: 1. Vertical section of 107 day foetus. 2. Parallel section of 107 day foetus. 3. Vertical section of 120 day foetus. 4. Parallel section of 120 day foetus. 5. Vertical section of 1 day skin.

differentiation, and their accessory structures have appeared. In another week or two, the anlagen of the secondary follicles have grown and expanded enough in the dermis to be seen at or above the level of the sebaceous glands of the primary follicles (figure 5, no. 2). The primary

follicles of the 4-month foetus are well developed and many of their fibers pierce the skin at the above age (figure 5, no. 2). For development of both primary and secondary follicles consult figure 5, no. 5, and figures 6, 7, and 8; for the depth of follicular penetration of the

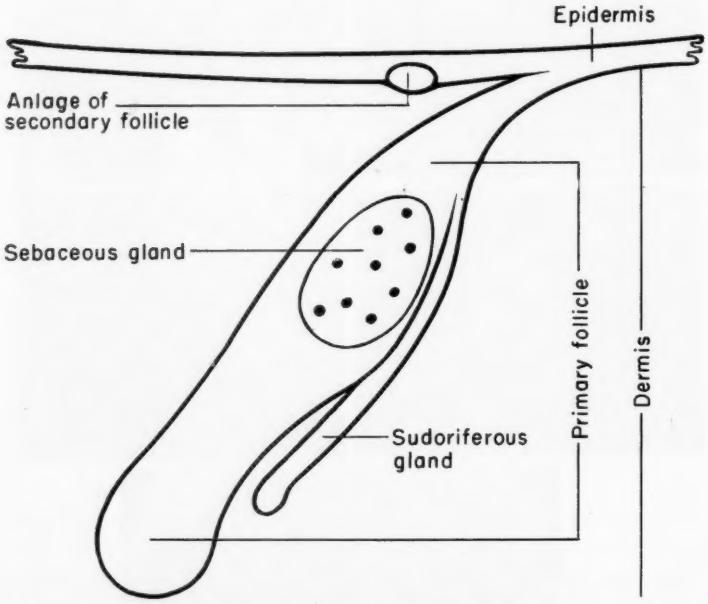

Figure 5a. Diagrammatic presentation of longitudinal section of primary follicle and anlage of a secondary follicle in a 90 to 100 day foetal skin.

undifferentiated to the fully formed foetal and for post-natal follicles, refer to figures 1, 2, 3, and 4.

The above measurements show that ventral follicles in mature animals reach a depth of 1.25 mm., dorsal follicles about 2 to 2.50 mm.

Skin Thickness as Related to Follicular Growth

The dermis is not differentiated into all its component elements until the foetuses are 3½ to 4 months old. Nevertheless, even in the early pre-follicular stages the skin can be delineated from the tissue underlying it by a relatively close disposition of the mesenchymal cells. In the 70-day foetus, when the primary anlagen have appeared in the epidermis—an orientation of the cells of this otherwise undifferentiated

dermis along the surface of the body can be noticed. In Karakul sheep there is no continuous muscle layer separating the dermis from the underlying layer as is the case in some other animals. In the present observations the dermis is delineated from the subcutis, a region where

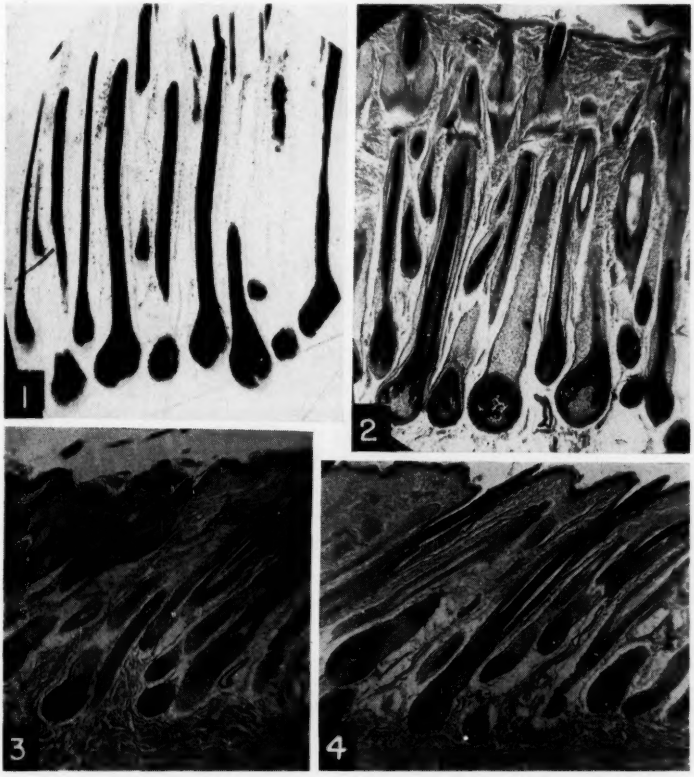

Figure 6. Vertical sections of mid-dorsal area of skin: 1. 5 day lamb. 2. 10 day lamb. Vertical sections of mid-ventral area of skin: 3. 5 day lamb. 4. 10 day lamb.

larger blood vessels are present and where looser connective tissues prevail.

As may be seen in figures 1 through 4, there is a rather close parallelism between the increasing thickness of the skin and the depth of follicular penetration. In foetal skins the dorsal dermis increases in thickness about 6.5 times from 70 days to birth, and the ventral dermis about 5

times during the same period. But the change in thickness of the dermis of post-natal sheep is less spectacular. For example, the dermis of a

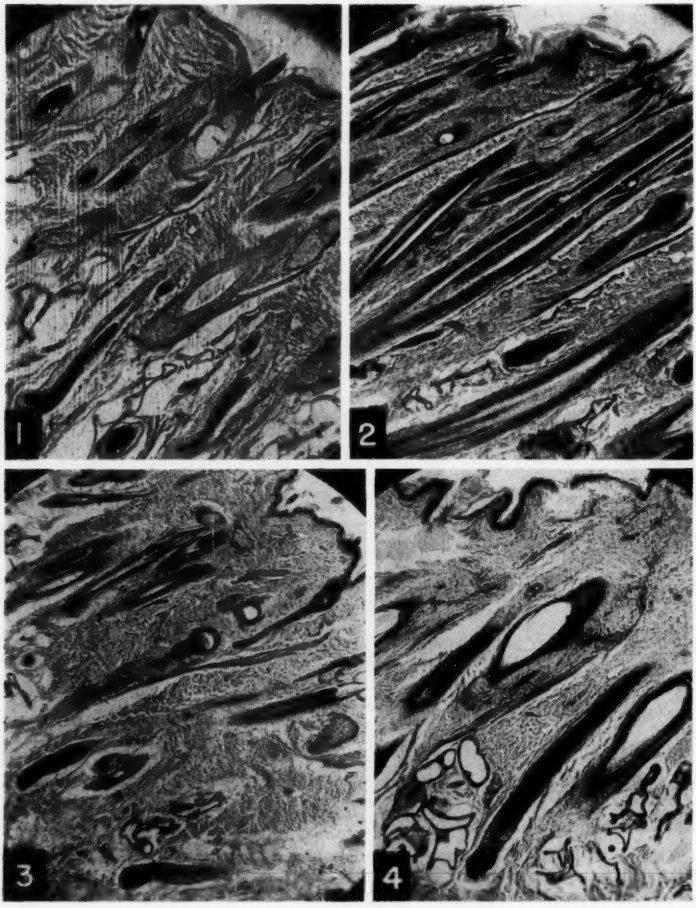

Figure 7. Vertical sections of mid-dorsal area of skin of different ages: 1. 1 year. 2. 2 years. 3. 4 years. 4. 6 years.

1-day lamb in the dorsal region is only one-half as thick as that of a 6-year-old sheep, and there is still less difference in the ventral samples, where a new-born lamb's dermis is less than half as thick as that of

an older sheep. The average ventral dermis of sheep barely reaches 2 mm., while the heaviest dorsal sample measured slightly over 5 mm.

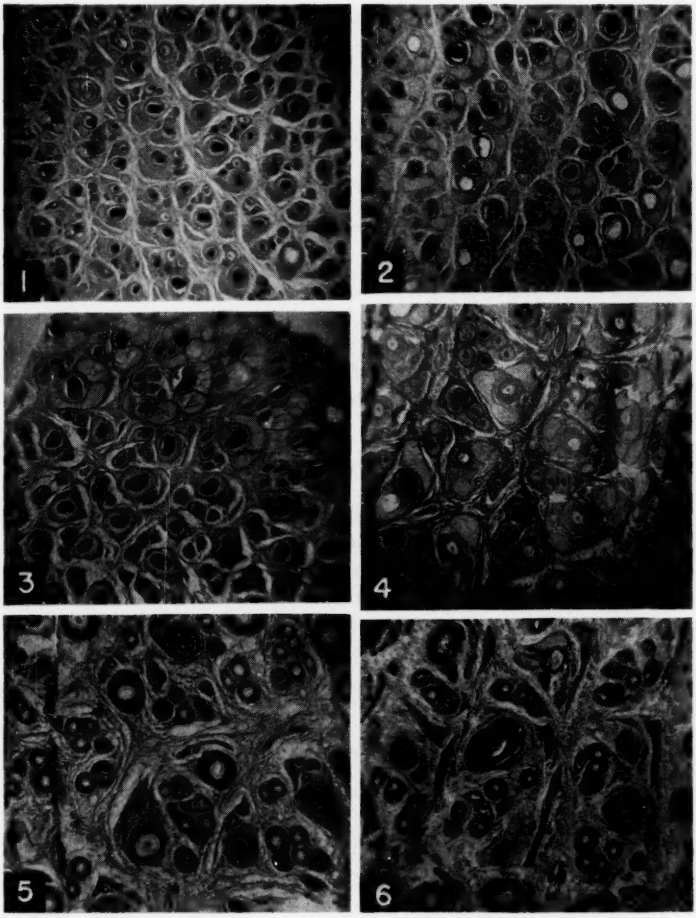

Figure 8. Parallel sections of mid-dorsal area of skin of different ages: 1. 1 day. 2. 10 day. 3. 30 day. 4. 2 year. 5. 4 year. 6. 6 year.

Direction and Form of Karakul Follicles

Students of Karakul skins seem to have overlooked the logical necessity of examining follicles in regions of poorest as well as of best curl

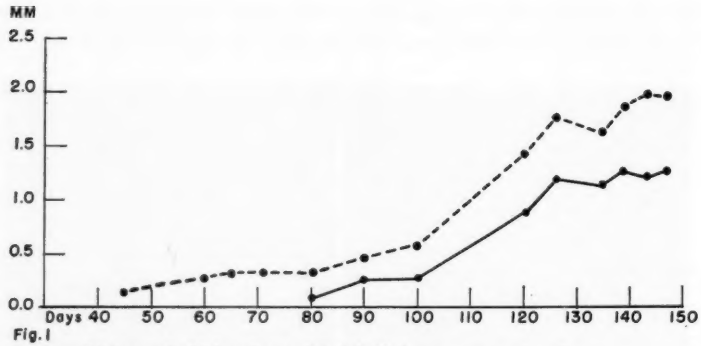

Fig. 1
Average thickness of mid-dorsal area of foetal skin in mm ---●---●---●
Maximum depth of penetration of primary follicles in mid-dorsal foetal skin in mm ●——●——●

formation, and the ventral skin was apparently not studied. Somewhat slanting follicles found in the rump and shoulder in animals past their best curl stage were supposed to be representative of all Karakul follicles. The present observations show that follicles forming the most desirable type of curl of the Karakul dorsum of the 1 to 10-day lamb are straight.

The differentiating follicle in the foetus is oblique in direction and this is more evident in the ventral area than in the dorsal area. However, the follicles in the dorsum undergo a gradual straightening at the end of the gestation period resulting in follicles which stand at right angles to the skin surface (figure 5, no. 5; figure 6, no. 1 through 4; and figure 7, no. 1 through 4). It may then be stated that the best Karakul curls are produced by fibers situated in straight follicles, which develop as

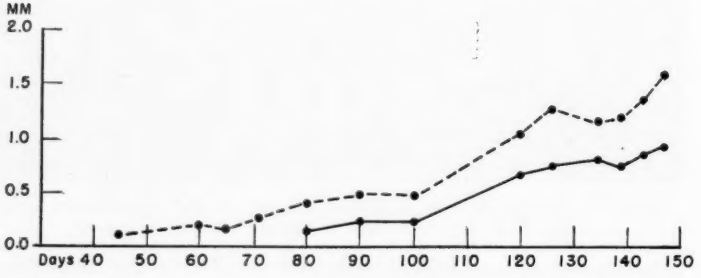

Fig. 2
Average thickness of mid-ventral area of foetal skin in mm ---●---●---●
Maximum depth of penetration of primary follicles in mid-ventral foetal skin in mm ●——●

close to one another as possible. With the gradual loosening of the curl shortly after birth, the follicles again begin to slant on the dorsum. In yearlings and mature sheep the dorsal follicles are slanting, but they never reach the extremely bent posture of older ventral follicles.

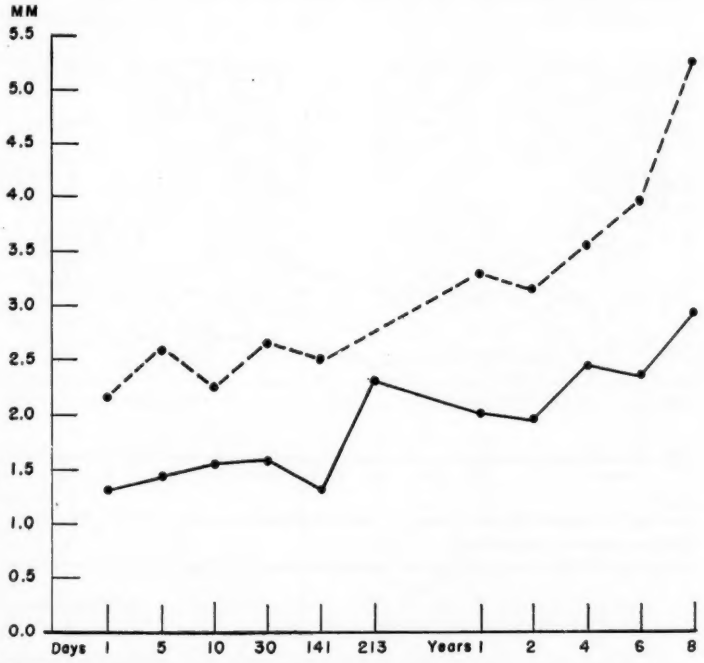

Fig. 3
Average thickness of mid-dorsal area of lamb and sheep skin in mm
Maximum depth of penetration of primary follicles in mid-dorsal area of lamb and sheep skin in mm

Proportion Between the Primary and Secondary Follicles

In the foetus the secondary follicles seem to undergo a growth less uniform than the primary ones. Parallel sections of foetal skins 15 weeks old show that usually only one secondary follicle (figure 5, no. 2), has grown down to the level of the sebaceous gland of its neighboring primary follicle. In some 2 or more weeks, that is, in foetuses of about 4 months, the relation between the primary and secondary follicles at the above level is as often 1 to 1 as 1 to 3 or 1 to 2, while in a lamb a

few days after birth most primary follicles of the dorsal sections (figure 8, nos. 1 and 2) are accompanied by no less than 2, but usually more secondary follicles. In a month-old lamb, secondary follicles have elongated enough to present a picture ranging from 2 to 5 secondary follicles to 1 primary one. That proportion remains pretty constant throughout life.

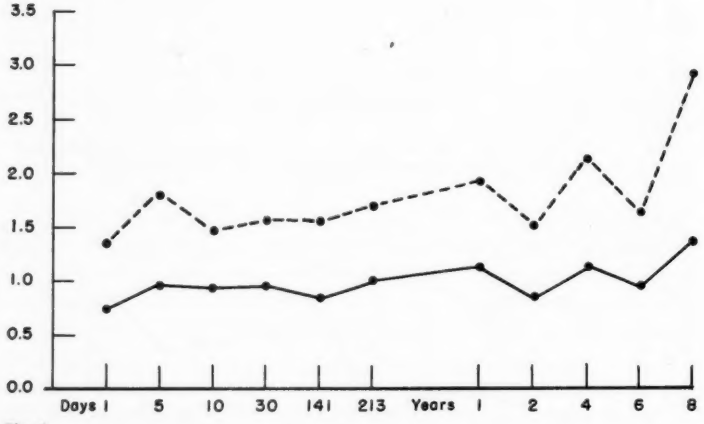

Fig. 4
Average thickness of mid-ventral area of lamb and sheep skin in mm
Maximum depth of penetration of primary follicles in mid-ventral area of lamb and sheep skin in mm

Number of Follicles Per Unit Area of Skin

As stated previously, no new follicular anlagen are produced in normal skins of post-natal Karakul sheep. The final proportion between the primary and secondary follicles becomes established within the first month of post-foetal life. Thus, as the follicular population remains constant throughout life while the body surface increases, a decrease of the number of follicles per unit area would be expected to take place. That is just what happens; the follicular groups vary in number in inverse proportion to the increasing surface of the growing animal. A group is considered to consist of a primary follicle, its accessory structures, and the secondary follicles in its immediate vicinity. As competition for space is quite clearly evident in the foetal and post-natal skins, considerable displacement naturally occurs. The secondary folli-

cles of one group may be squeezed towards a neighboring group, and particularly in older animals other factors like changes in texture and consistency of the dermis would tend to alter the original location of the more slender secondary follicles. It has therefore been found practical to use the primary follicle as the center of a basic unit of a group, consult diagram figure 9.

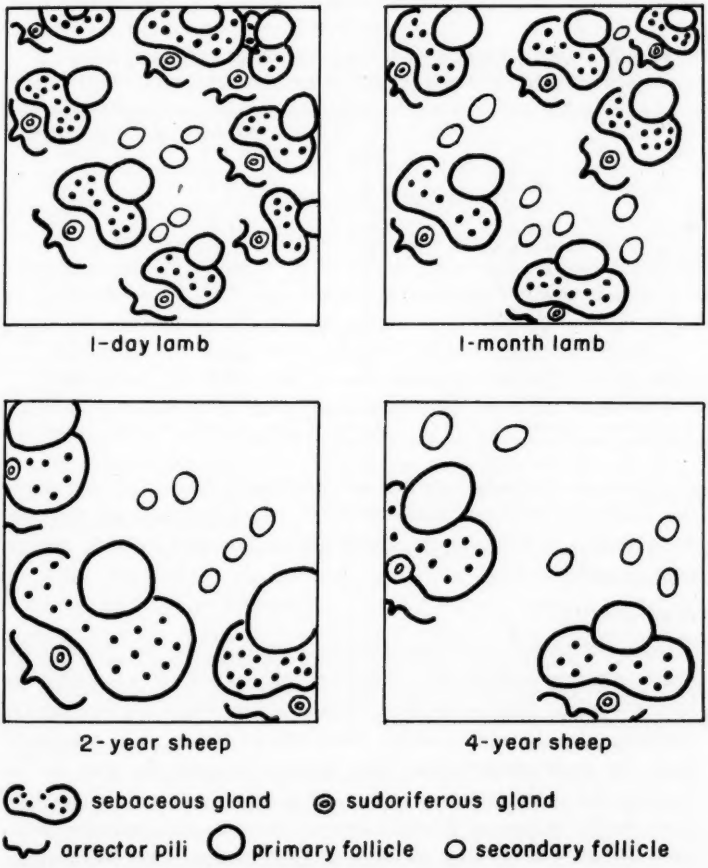

Figure 9. Diagrammatic presentation of cross sections of follicles in the region of sebaceous glands showing the number of primary follicles at different ages per unit area of dorsal skin.

The averages of follicular groups counted are very close to expectations; namely, a 4-year sheep's body surface is 3.88 times larger than that of a 1-day lamb, and while the 1-day lamb shows 7 to 8 follicular groups, for a given area, a 4-year sheep shows only 2 for the same area. Note that the body surface of a 2-year-old sheep is 2.23 times larger than that of a 30-day lamb, and while the latter shows 6 follicular groups, the former shows only 3 on the same area. If one considers that

TABLE 1. AGE, NUMBER, SEX, WEIGHT, AND BODY SURFACE OF SOME LAMBS AND SHEEP USED IN THE PRESENT STUDY

Age	No.	Sex	Weight in lb.	Body surface in m^2
1 d.	699-K	m.	9.1	0.28
10 d.	698-K	f.	12.7	0.33
30 d.	206-KN	m.	25.2	0.47
141 d.	191-KN	m.	55.0	0.69
1 yr.	457-KB	f.	91.0	1.01
2 yr.	395-KC	f.	100.0	1.05
4 yr.	114-KN	f.	111.0	1.11
6 yr.	295-KC	f.	154.0	1.33
6 yr.	541-K	m.	162.0	1.37
8 yr.	249-KC	f.	127.0	1.20

Photomicrographs of parallel sections of the above 1, 10, 30 day, and 2, 4, and 6 year (female) skins are presented in figure 8; diagrams of cross sections of follicles of the above 1 and 30 day, and 2 and 4 year old skins are presented in figure 9.

the surface of the body increases from birth to maturity 3½ to 4 times, the number of follicles should decrease correspondingly, as they do, from about 7 to 8 to only 2, consult Fig. 9. For data on body surface refer to table 1.

Fiber Density

Inasmuch as Karakul sheep produce only one fiber per follicle, the numbers and proportions established for follicles will be potentially the same for fibers. Thus, with the exception of the possibility of irregular shedding, or pathologic changes, there will be from 2 to 5 secondary fibers for each primary one. Since density remains the goal of the breeders and if the follicles of other sheep follow a pattern of development similar to that of Karakul sheep, it is clear that increased postnatal growth will offer no solution. It must, however, be also realized that in Karakul sheep the foetal follicles seem to fill the available space to capacity.

On the basis of the above information—the problem of a denser

population could probably be approached by one of the following methods:

(1) Selection of breeding stock from individuals with particularly dense populations of fibers. Sheep with dense fleeces would have evidently originated from foetuses with unusually large body surfaces at the critical period of follicular initiation which is from 60 to 65 days.

(2) Breeding of animals selected for finer fibers and with the same capacity for filling the available skin area.

(3) Attempts to accelerate foetal growth just preceding the second half of gestation when follicular anlagen make their appearance. This could perhaps be achieved by means of nutrition including vitamins or growth substances.

Summary and Conclusions

Two basic generations of fiber follicles are produced by Karakul sheep foetuses, and no further generations are initiated during post-natal life.

Because of the fact that no new follicles are initiated during post-natal stages—forcing sheep to produce larger body surfaces after birth in the hope of obtaining denser fleeces—cannot be recommended.

The problem of developing denser fleeces can be approached by the customary method of selecting breeding stock from the most desirable individuals for the character in question; it can be also attacked by attempts to accelerate foetal growth just preceding the second half of gestation, thus taking advantage of our knowledge of when follicular anlagen are being initiated.

There is a definite sequence of development of the two types of follicles characteristic of Karakul sheep. The anlagen of the primary follicles develop in foetuses at about 70 days, while those of the secondary follicles appear 3 to 4½ weeks later. Thus the primary and the secondary follicles are established during foetal life.

As the follicular population is constant throughout life, the number of follicles per unit area will vary in inverse proportion to the increasing surface of the body.

There is an almost direct relationship between the thickness of the skin and the depth of follicular penetration; both increase with age.

The epidermis proper remains a thin sheath composed of hardly more than three layers of cells from birth to old age.

Since no more than one fiber is produced by either the primary or the secondary follicle of Karakul sheep, the proportion between the primary and the secondary fibers will be the same as for follicles, that

is 1 to about 2 to 5. Irregularities caused by shedding or pathologic skin conditions are excluded.

The first keratinization of the primary fiber occurs in foetuses about 15½ weeks old.

Fibers which produce the most desirable type of curl in Karakul sheep, for example those in the dorsal skin of animals 1 to 10 days old, are found in straight follicles distributed in almost juxtaposition to one another. On the other hand, ventral fibers, which produce the poorest curls, are in slanting follicles of sparser distribution.

Literature Cited

Carter, H. B. 1939. A histologic technique for the estimation of follicle population per unit area of skin in the sheep. Jour. Council Scien. & Ind. Res., Australia, 12:250.

Duerden, J. E. and M. J. Ritchie. 1924. Development of the Merino Wool Fiber. South African Jour. Sci. 21:480.

Herre, W. and I. Rabes. 1937. Studien an der Haut des Karakulschafes. Zeitschr. mikr. anat. Forschung, 42:525.

Herre, W. and H. Wigger. 1939. Die Lockenbildung der Saügetiere. Kühn Archiv. 52:233.

Leshchinskaia, E. M. 1952. Seasonal changes of the skin of mammalian animals. Jour. Zool. 31:434. Moscow. (In Russian.)

Margolena, Lubow A. and Ethel H. Dolnick. 1953. Cell division in the epidermis of Karakul sheep in relation to age and the development of hair follicles. Va. Jour. Sci. 4:71 (abstract).

Nikolskii, N. F. 1932. The formation of curl in Karakul pelts. Bull. zool. techn. and breeding station of the State Experiment Inst. Chapli. No. 7. (In Russian)

Ritzman, E. G. and N. F. Colovos. 1930. Surface area of sheep. Circular 32, New Hampshire Agricultural Experiment Station.

ABSORPTION AND TISSUE DISTRIBUTION OF RADIOZINC IN STEERS FED HIGH-ZINC RATIONS [1]

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IT has been shown by Bertrand and Bhattacherjee (1934) and by Elvehjem and associates (1934, 1935) that zinc is a necessary constituent of the diet for the normal growth of mice and rats. Excess dietary zinc was reported by Sutton and Nelson (1937) to produce anemia, subnormal growth and reproductive failure in rats. Gray and Ellis (1950) noted that the inhibitory effect of molybdenum on growth of rats was aggravated by the addition of zinc to the diet, but observed no retardation of growth upon addition of zinc alone. Scott and Fisher (1938) observed weight losses in mature cats fed on a high zinc diet. McCance and Widdowson (1942) observed that in humans, excess dietary zinc was excreted in the feces, with no corresponding increase in urinary zinc excretion. The excretion and tissue distribution of injected radioactive zinc^{65} has been studied in dogs and mice by Sheline *et al.* (1943). The radioisotope has also been employed in studies of the relationship of zinc to the pancreas (Montgomery and co-workers, 1943; Heath and Liquier-Milward, 1950; Lowry, Baldwin and Harrington, 1954) and blood plasma and cells (Ebaugh and Ross, 1952; Tupper and Watts, 1952).

The present investigation was undertaken to study the movement of zinc through the animal body and to observe possible effects of excess dietary zinc on the condition of cattle and on the absorption and distribution of radioactive zinc^{65} administered orally and intravenously to young growing steers.

Experimental

Four purebred Jersey steers were maintained from the age of one month on a ration of grain (from ½ pound at one month to 6 pounds

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at one year), 1 pound cottonseed oil meal and prairie hay according to the calculated requirements of the animal. Zinc carbonate was added at levels of 1000 ppm. zinc to the total ration of two of the steers and 50 ppm. to the total ration of the other two (control) steers. At 13 and 18 months of age, respectively, the high zinc and control animals were placed in metabolism units for the quantitative separation and collection of urine and feces. After the animals had become adjusted to the unit and were on a constant feed intake they were given orally or intravenously (Hansard *et al.*, 1951) a single dose of $zinc^{65}$, and concurrent six-day chemical and radioisotope balance studies were started.[5]

Blood, feces and urine samples were taken at regular intervals for assay. At the end of 6 days, the animals were slaughtered. Tissue samples were carefully selected, weighed and ashed. The ash was dissolved in acid and the solution made to volume. Radiozinc measurements were made with dipping type Geiger-Muller Counters and conventional scalers. Total zinc content of selected tissues was determined by means of the Leeds and Northrup polarograph after extraction of zinc by the dithizone procedure of Menzel and Jackson (1940).

Results and Discussion

Absorption and Excretion of Zinc

The results of the 6-day balance studies indicated that only 3–10 percent of the dietary zinc was absorbed from the gastrointestinal tract. There were no gross symptoms of toxic effects observed in the two animals receiving the high levels of dietary zinc, and although their weight gains were slightly lower than those of the controls, the difference was not statistically significant. The high-zinc animals at slaughter weighed 427 and 480 pounds at 378 and 409 days of age, while the control steers weighed 540 and 560 pounds at 505 and 539 days of age.

Likewise, there were no significant effects noted on $zinc^{65}$ excretion that could be attributed to the difference in dietary zinc intake. Following oral administration, however, the high-zinc steer excreted slightly more of the isotope in the feces (73.2%) than the control animal (65.8%). Urinary excretion accounted for 0.24 and 0.39 percent of the administered dose. Following intravenous injection of zinc, fecal values were 20.6 and 20.7 percent and urine values were 0.33 and 0.13 percent of the injected dose for the high-zinc and control animals, respectively.

[5] The labeled $zinc^{65}$ was obtained from the Oak Ridge National Laboratory, Oak Ridge, Tennessee, after allocation by the Isotope Division of the Atomic Energy Commission.

These differences between animals are within the expected range of individual variation and would indicate little effect of dietary zinc intake upon $zinc^{65}$ behavior in this species. Sheline *et al.* (1943) have reported that in mice 50 percent of intravenously injected $zinc^{65}$ appeared in the feces in 7 days, and in dogs 25 percent appeared in the feces in 15 days, less than 5 percent being excreted in the urine during these periods.

Following oral administration of radiozinc, peak excretion of the isotope in the feces occurred during the 24- to 48-hour interval. After intravenous administration there was no such clearly defined peak, although radiozinc was excreted at a slightly higher rate from 24 to 120 hours after dosing than during the remainder of the dosage-to-sacrifice interval.

$Zinc^{65}$ in the Blood

In order to study the movement and exchange of radiozinc between blood and tissues, blood samples were drawn at frequent intervals during the 6-day period. Analysis of 18 blood samples from the steers dosed orally indicated that $zinc^{65}$ entering the blood from the gastrointestinal tract never reached concentrations sufficiently high to measure. Following intravenous administration of the isotope, however, the level of $zinc^{65}$ decreased rapidly from 100 percent of the dose in the total blood at 3 minutes to 10 percent after 4 hours. Total blood was calculated as 7 percent of body weight (Hansard *et al.*, 1953). Equilibrium between blood and tissue appeared to have been reached after about 10 hours, with approximately 3 percent of the administered dose present in the blood. Ebaugh and Ross (1952) reported that 11 percent of the $zinc^{65}$ dose remained in the plasma of man at 30 minutes after intravenous administration.

Since zinc is known to be a constituent of the enzyme carbonic anhydrase, found in red blood cells, an attempt was made to follow the *in vivo* uptake of radioactive zinc by the red cells. Packed cell volume was determined by centrifuging the blood samples, and blood cell $zinc^{65}$ activity calculated by subtracting from the counts per minute in a given volume of whole blood the counts found in the volume of plasma present in this volume of blood according to the cell-plasma ratio for the sample. Figure 1 shows the rate of disappearance and the partition of $zinc^{65}$ between bovine plasma and cells as a function of time following intravenous isotope administration. $Zinc^{65}$ values are expressed as counts per minute in the plasma and cell fractions making

up 100 ml. of whole blood. The values at 3 and 10 minutes represent averages of those obtained with blood from both steers; the remaining points are based on single values for individual samples drawn alternately from the two animals.

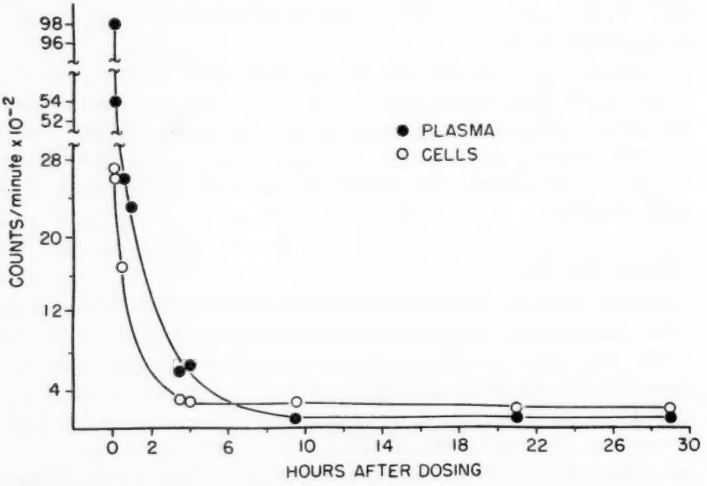

Figure 1. The disappearance of zinc^{65} from plasma and red cells of bovine blood following intravenous administration. (Counts per minute per 100 ml. whole blood equivalent.)

In the samples drawn 3 minutes after isotope administration, 22 percent of the radioactive zinc in the blood was found in the cell fraction and 78 percent in the plasma. A steady increase in the cell-to-plasma zinc^{65} ratio was noted during the next several hours, equilibrium between the two fractions being reached about 6 hours after dosage. Thereafter, with plasma zinc^{65} activity showing a slightly more rapid decrease than that of the cells, cellular activity predominated. These findings indicate that the red cell wall is permeable to the zinc cation, a concept in keeping with the essential nature of this element with respect to the red blood cell enzyme carbonic anhydrase.

Keilin and Mann (1939) found the zinc content of carbonic anhydrase prepared from beef corpuscles to be directly proportional to the enzyme activity, and were of the opinion that all the zinc present in the erythrocytes was combined with carbonic anhydrase; they concluded that this enzyme is a zinc-protein complex. Tupper, Watts and

Wormal (1952) corroborated the fact that zinc in carbonic anhydrase is firmly bound to the protein, noting that, *in vitro* at least, little if any of the zinc^{65} used in labeling red cells became firmly attached to the carbonic anhydrase. Berfenstam (1952) observed that intravenous injections of zinc raised the zinc content of the erythrocytes without a concomitant increase in carbonic anhydrase activity. Whether or not in the present study the zinc^{65}, which apparently crossed the red cell membrane, actually exchanged with the zinc ions already attached to the carbonic anhydrase is problematical.

Tissue Distribution of Zinc^{65}

Excretion data previously presented indicate that retention of zinc^{65} averaged 30 and 79 percent for oral and intravenous administration, respectively. This was reflected in the zinc^{65} content of the tissue in that the intravenous values were from two to three times the corresponding oral values. As discussed earlier, the dietary zinc level did not appear to influence the behavior of radiozinc. Therefore, in order to eliminate effects due to differences in methods of administration, absorption from the tract or body weight, all tissue distribution values for zinc^{65} have been calculated on the basis of percentage of *retained* zinc^{65} dose, and corrected to a standard body weight of 500 pounds. Thus the values tabulated in the second column of table 1 for individual tissues reflect the handling by the body of that radiozinc made available to it.

Since there was no apparent influence of dietary zinc levels either upon total tissue zinc or upon the behavior of the administered radiozinc, the summary of the distribution of zinc^{65} in selected tissues 6 days after oral and intravenous administration to all steers is presented in table 1. Concentration values for total zinc are expressed as mg. of zinc per kg. fresh weight, and tissues are arranged in decreasing order of concentration.

It is of particular interest that the pituitary contained the highest level of zinc and values for the incisors ranked along with the soft tissue values. The blood value of 8.8 mg. per kg. is in close agreement with the findings of Vallee and Gibson (1948). Scott and Fisher (1938) noted that in cats fed excess zinc at a slightly lower level per kg. of body weight than that received by these steers, zinc concentrations in the pancreas and liver were 480 and 900 micrograms per gram, respectively, as compared with 72 and 58 micrograms in the controls. This dietary influence was not noted for ruminants in this study where excess zinc was fed for more than a year.

The accumulation of radiozinc, in general, was highest in the soft tissues. The pancreas, liver, pituitary, kidney and adrenals showed the highest concentration of retained dose. However, the turnover rate as shown by specific activity (rapidity of uptake and loss) was highest in the liver, adrenals and pancreas. In decreasing order of concentration, the gastrointestinal tract and contents, muscle, spleen, hide and hair, brain, thymus, lungs, heart and spinal cord contained from .03 to 0.2 percent of the absorbed dose per kg. of fresh tissue. The bile and bone marrow contained no radiozinc after 6 days.

TABLE 1. SUMMARY OF ZINC^{65} DISTRIBUTION IN YEARLING CATTLE AT 6 DAYS AFTER ADMINISTRATION

Tissue	Mg. Zinc per Kg. Fresh Weight	Percentage of Zn^{65} Dose Retained per Kg. Fresh Tissue	Specific Activity[1]
Pituitary	247	0.98	4.0
Incisors	130	.17	1.3
Pancreas	125	1.58	12.6
Kidney	121	.86	7.1
Adrenals	55	.75	13.6
Rib Sternal End	45	.80	17.8
Liver	33	1.20	36.3
Rib Shaft	28	.15	5.4
Mandible Angle	25	.35	14.0
Whole Blood	8.8	.04	4.5

[1] Specific activity expressed as percent of *retained* zinc^{65} dose per milligram zinc X 10^3.

Radiozinc accumulation in bones more or less paralleled their metabolic activity, probably reflecting the blood supply to the particular areas. Trabecular bone retained more zinc^{65} than cortical bone and the turnover rate, as shown by the specific activity values, was higher for the sternal end of the rib than for the rib shaft; turnover rate was higher for the mandible than for the metatarsus shaft and was lowest for the incisor teeth.

The pancreas and liver showed the highest concentrations of radiozinc in all 4 animals. This finding with respect to the pancreas bears out the close chemical relationship between insulin and zinc. Sheline and co-workers (1943) found that the tissues showing the greatest turnover following intravenous administration of zinc^{65} to dogs were the liver, pancreas and kidney. Slowest turnover occurred in red blood cells, brain, skeletal muscle and skin. Heath and Liquier-Milward (1950) observed that zinc^{65} was present in relatively large amounts in the pancreas as well as the liver, spleen and kidney of humans

following intramuscular injection of the isotope, and zinc^{65} was found by Montgomery *et al.* (1943) to be eliminated in the pancreatic juice of the dog. Lowry, Baldwin and Harrington (1954) noted that in alloxan-diabetic rats the uptake of zinc^{65} by the pancreas was considerably lower than in normal rats.

Summary

Radiochemical procedures have been employed to study the movement of zinc through the body of four yearling steers. Absorption and tissue distribution in two steers maintained for more than a year on 1000 ppm. zinc were compared with control animals following oral and intravenous administration of the radioisotope.

Concurrent chemical and radiozinc balance studies indicated that most of the zinc was excreted in the feces and was not absorbed. Those steers dosed orally with zinc^{65} excreted 72.2 and 65.8 percent in the feces and 0.24 and 0.39 percent in the urine on the high-zinc and control rations, respectively. Following intravenous administration, 20.6 and 0.25 percent of the dose was excreted in the feces and urine, respectively, with no differences attributable to dietary zinc level noted.

The rate of disappearance of zinc^{65} from the blood following intravenous administration resulted in the attainment of equilibrium between blood and soft tissue in 10 hours. The partition between plasma and cells presented as a function of time indicated that equilibrium between the two fractions occurred at about 6 hours, after which activity in the red cells predominated.

The accumulation of *retained* radiozinc was, in general, highest in the soft tissue, the pancreas, liver, pituitary, kidney and adrenals having highest concentration. Turnover rate was highest for the liver and lowest for the incisor teeth.

The accumulation and turnover of zinc in the various bones paralleled their metabolic activity.

Literature Cited

Berfenstam, R. 1952. Studies on carbonic anhydrase activity in children. Acta paediat. 41:32.

Bertrand, G. and R. C. Bhattacherjee. 1934. The combined action of zinc and vitamins in the feeding of animals. Compt. Rend. Acad. 198:1823.

Ebaugh, F. G. and J. F. Ross. 1952. Radioactive zinc metabolism in patients with neoplastic diseases. Fed. Proc. 11:413.

Gray, L. F. and G. H. Ellis. 1950. Some interrelationships of copper, molybdenum, zinc, and lead in the nutrition of the rat. Jour. Nutr. 40:441.

Hansard, Sam L., C. L. Comar and M. P. Plumlee. 1951. Radioisotope Procedures with farm animals. I. Nucleonics 9:13.

Hansard, Sam. L., C. L. Comar and M. P. Plumlee. 1951. Radioisotope Procedures with farm animals. II Nucleonics 9:38.

Hansard, Sam L., W. O. Butler, C. L. Comar and C. S. Hobbs. 1953. Blood volume of farm animals. JOURNAL OF ANIMAL SCIENCE 12:402.

Heath, J. C. and J. Liquier-Milward. 1950. Distribution and function of zinc in normal and malignant tissues. I. Uptake and distribution of radioactive zinc^{65}. Biochem. Biophys. Acta 5:404.

Keilin, D. and T. Mann. 1939. Carbonic anhydrase. Nature 144:442.

Lowry, J. R., R. R. Baldwin, and R. V. Harrington. 1954. Uptake of radiozinc by normal and diabetic rat pancreas. Science 119:219.

McCance, R. A. and E. M. Widdowson. 1942. The absorption and excretion of zinc. Biochem. Jour. 36:692.

Menzel, R. G. and M. L. Jackson. 1951. Determination of copper and zinc in soils or plants. Anal. Chem. 23:1861.

Montgomery, M. L., G. E. Sheline and I. L. Chaikoff. 1943. Elimination of administered zinc in pancreatic juice, duodenal juice and bile of the dog as measured by its radioactive isotope (Zn^{65}). J. Exp. Med. 78:151.

Scott, D. A. and A. M. Fisher. 1938. The pancreas and liver of normal and zinc-fed cats. Am. Jour. Physiol. 121:253.

Sheline, G. E., I. L. Chaikoff, H. G. Jones, and M. L. Montgomery. 1943. Studies on the metabolism of zinc with the aid of its radioactive isotope. I. Excretion of administered zinc in urine and feces. Jour. Biol. Chem. 147:409.

Sheline, G. E., I. L. Chaikoff, H. B. Jones and M. L. Montgomery. 1943. Studies on the metabolism of zinc with the aid of its radioactive isotope. II. The distribution of administered radioactive zinc in the tissue of mice and dogs. Jour. Biol. Chem. 149:139.

Stirn, F. E., C. A. Elvehjem and E. B. Hart. 1935. The indispensability of zinc in the nutrition of the rat. Jour. Biol. Chem. 109:347.

Sutton, W. R. and V. E. Nelson. 1937. Studies on zinc. Proc. Soc. Exp. Biol. Med. 36:211.

Todd, W. R., C. A. Elvehjem and E. B. Hart. 1934. Zinc in the nutrition of the rat. Am. Jour. Physiol. 107:146.

Tupper, R., R. W. E. Watts and A. Wormal. 1952. Some experiments on the zinc in carbonic anhydrase. Biochem. Jour. 50:429.

Vallee, B. L. and J. G. Gibson. 1948. The zinc content of normal human whole blood, plasma, leucocytes and erythrocytes. Jour. Biol. Chem. 176:446.

THE EFFECT OF TRACE MINERALS ON GROWTH PERFORMANCE AND VITAMIN B_{12} SYNTHESIS OF STEERS

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THE mineral composition of roughages and pasture plants has a significant effect on their feeding value for ruminant animals. This relationship has been reviewed by Bohstedt (1942), Mitchell (1947), and Russell (1944), particularly with reference to calcium and phosphorus. Under feed lot conditions, Klosterman *et al.* (1953) observed that steers fed poor quality mature hay as part of a typical fattening ration responded to the ash of alfalfa or the ash of dried molasses fermentation solubles or a trace mineral mixture composed of iron, copper, cobalt, manganese, and zinc. Steers receiving these supplements gained significantly faster and were more efficient in the conversion of their rations. A part of this effect appears to be elicited through improved rumen function. Swift *et al.* (1951) found that the addition of alfalfa ash increased the consumption of a purified-type lamb ration. Burroughs *et al.* (1950) showed that the digestibility of a corn cob ration was improved by the addition of alfalfa ash. Tillman *et al.* (1954) fed alfalfa ash to sheep on a prairie hay ration but there was no increase in apparent digestibility of the hay. These results plus the report by Burroughs *et al.* (1951), showing the stimulatory effect of ashed residues of molasses etc. on rumen microorganisms *in vitro,* has suggested the need for further study on the importance of trace mineral elements on ruminant nutrition.

The results to be presented herein are concerned with the effect of alfalfa ash or trace mineral elements on the rate of gain, feed efficiency, and ruminal vitamin B_{12} synthesis of steers fed a fattening type ration which contained mature timothy hay. Preliminary reports of these studies have been presented previously (Bentley *et al.* 1952, 1953).

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Experimental

Steer Feeding Experiments

In each of two trials, 12 good quality grade Hereford steers were stanchioned individually in stalls equipped with cloth-laminated rubber mats (Musser Rubber Company, Akron, Ohio). A ration composed of ground ear corn, chopped poor-quality timothy hay, and 4.0 lb. of a supplement having the following percentage composition—urea, 5.00; calcium hydrogen phosphate (USP grade), 1.43; calcium carbonate (USP), 1.05; iodized salt, 1.24; cerelose, 42.5; and ground ear corn, 48.78—was hand, full fed. This supplement supplied nitrogen, phosphorus, calcium, and salt to balance the deficiencies of the corn-poor hay components of the ration, according to National Research Council recommendations (1950). Vitamins A and D were supplied separately by a ground ear corn-feeding oil (6000 A-300 D per gm.) mixture which was fed to supply about 3000 I.U. of vitamin A per 100 lb. body weight daily. Daily feed consumption records were kept and the steers were weighed at two-week intervals. Water was provided at all times.

Urea (Du Pont's "262" or crystalline urea) supplied an average of 40 percent of the total nitrogen intake, and the ration as eaten contained from 11 to 12 percent crude protein. It required about one month for the steer calves to accept fully the supplement portion of the ration and to become adjusted to the stanchions in which they were kept.

The mature, late-cut timothy hay used in these studies was similar to that used by Klosterman *et al.* (1953) and analyses of several samples indicated that it contained 5.5–6.5 percent crude protein, 4.0 percent ash; 37 percent crude fiber (A.O.A.C., 1950) or 71 percent by the Wilcox-Moxon (1949) method, and little or no carotene.

Alfalfa ash was prepared by burning leafy, green 2nd and 3rd cutting alfalfa hay (about 8% ash). The grey ash was fed at the rate of 75 gm. per day per steer mixed with the supplement portion of the ration.

The trace mineral mixture used in this study contained the following salts, gms. per 100 gms.: manganous chloride ($MnCl_2 \cdot 4H_2O$), 44.25; cobaltous chloride ($CoCl_2 \cdot 6H_2O$), 0.123; zinc chloride ($ZnCl_2$), 1.787; cupric acetate ($Cu(CH_3COO)_2 \cdot H_2O$), 4.81; and reduced electrolytic iron, 49.03. This mixture was dried at 105° C. for 18 hours and finely ground through a burr mill and, where indicated, each steer received 1.465 gm. daily of this mixture which supplied the following amounts of supplemental trace mineral elements: copper, 25 mg., and cobalt, 0.5 mg. (Russell, 1944); manganese, 200 mg. (Bent-

ley and Phillips, 1951); iron, 800 mg.; and zinc, 14 mg. (Burroughs, 1952).

Chemical and Microbiological Methods

Feed and liver samples were analyzed for cobalt by the method of Arthur *et al.* (1953).

The method of Ketzes *et al.* (1944) was used to determine iron and A.O.A.C. (1950) procedures were used for copper and manganese. Samples were prepared for copper and iron analyses by the methods described by Parks *et al.* (1943).

Water extracts of feces or macerated liver (1 gm. material in 100 ml. water) were analyzed for vitamin B_{12} microbiologically using *Lactobacillus leichmannii* ATCC No. 7830. The assay medium of Skeggs *et al.* (1950) was used with slight modifications. Thiomalic acid was replaced by ascorbic acid and fumaric acid, according to the procedure used by Thompson *et al.* (1950). In the preparation of liver and kidney samples sodium cyanide was used to protect the vitamin B_{12} activity.

Digestion Experiment Using Chromic Oxide Marker Technique

The chromic oxide (Cr_2O_3, C.P. reagent grade) was thoroughly mixed with the supplement portion of the ration (10 gm. per 4.0 lb.—the amount of supplement fed each steer daily). After mixing, 5 different samples of the chromic oxide-containing supplement were collected using a grain sampler and were combined to give a composite sample for analysis. The composite samples of feeds and dried feces were then analyzed for chromic oxide using the method of Bolin *et al.* (1952). Kjeldahl nitrogen determinations were carried out on oven-dried feed samples, whereas wet feces samples were analyzed to avoid nitrogen losses during drying. Crude fiber determinations were made on the dry and fat-free feed and feces, according to the A.O.A.C. (1950) procedures. Dried feces and feed were also analyzed for crude fiber using the method recently suggested by Wilcox and Moxon (1949).

The chromic oxide marker was fed for 7 days prior to the beginning of the 8 day collection period. A composite feces sample from each steer was prepared by getting samples of freshly voided feces (about 100–200 gms.) between 8 and 9 o'clock in the morning and between 3 and 4 o'clock in the afternoon on each of the 8 days (Kane *et al.* 1952). During the collection period the samples were refrigerated and a few drops of a 10 percent alcoholic solution of thymol was added

daily to prevent bacterial growth. At the end of the collection period, the total wet feces sample was thoroughly mixed and a portion removed for nitrogen analysis. The remainder of the sample was dried in a forced-air oven at about 60° C., ground in a Wiley mill, and then sampled for crude fiber and chromic oxide determinations.

Digestibility coefficients were calculated on a moisture, fat, and ash-free basis and, in general, procedures followed were similar to those reported by Kane *et al.* (1950) and Schürck *et al.* (1950).

Results

Feeding Experiments

The results of the first experiment (1952), which are presented in table 1, show that feeding either alfalfa ash or a mixture of 5 trace minerals significantly increased average daily gains. Feed efficiency was improved and ground ear corn consumption was increased about 25% but these differences were not significant. Iron supplementation had no effect on average daily gains, corn consumption, or feed efficiency. An iron supplement was used since an iron requirement for rumen microorganisms *in vitro* had been demonstrated by McNaught *et al.* (1950).

After the calves had been on the rations described in table 1 for 140 days, the first phase of the experiment was terminated. At this time the calves on "basal" or the "basal plus iron" diets appeared to be deficient in trace minerals, as indicated by poor growth and lack of appetite. These steers were then fed the basal ration supplemented with either copper, maganese, cobalt, or the trace mineral mixture. The results of an 84-day experiment are given in figure 1. For comparison, the average daily gain of each steer during the first phase of the experiment is also given. Copper, manganese, and cobalt were fed singly at the same level as they were supplied in the trace mineral mixture.

These steers deficient in trace mineral responded to either the trace mineral mixture or 0.5 mg. cobalt daily. There was some growth improvement shown by the steer fed the manganese supplement but the effect was not marked. Copper appeared to have no effect. Steers number 1 and 60, which were continued on the basal ration during the second phase of the experiment, showed a reduced rate of gain indicating that as the mineral deficiency became more severe, the rate of gain was further reduced. One of the steers on basal diet (No. 1), fed cobalt for 21 days, showed a marked improvement in appetite and gained 84 lb. This was probably largely fill but was indicative of improved appetite.

These preliminary results suggested that the first limiting trace mineral deficiency observed was the lack of cobalt. Vitamin B_{12} analyses of feces obtained from the steers deficient in trace minerals also suggested a lack of dietary cobalt for ruminal vitamin B_{12} synthesis. In

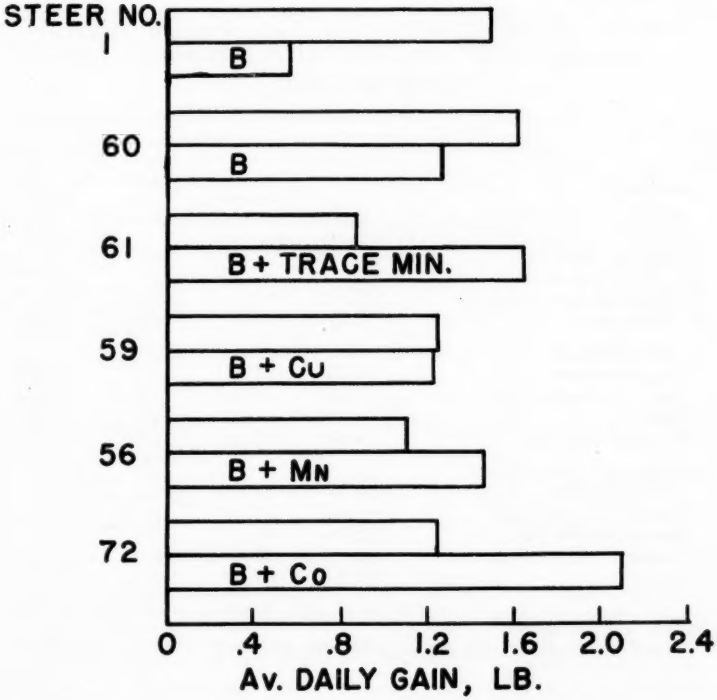

Figure 1. The upper bars in each pair indicates the gains of steers fed the "basal" (1, 60, and 61) or the "basal plus iron" (56, 59, and 72) rations during the first 140 day phase of the 1952 experiment. The gains obtained with the basal ration alone or supplemented as indicated during the second phase (84 days) of the experiment are shown by the lower bars of each pair.

1953, a second steer experiment was conducted to evaluate this observation, the results of which are given in table 2.

As in the previous experiment, adding trace minerals to a ground ear corn-mature timothy hay-urea-minerals basal ration increased the average daily gains of steer calves significantly and, in addition, ground ear corn consumption was 33% greater for the mineral-fed-steers (sig-

nificant at 5% level). When cobalt was fed alone, average daily gains increased 0.26 lb. which approached significance at the 5% level. However, ground ear corn consumption was increased 32.5% which was significant.

TABLE 1. THE EFFECT OF IRON, A TRACE MINERAL MIXTURE AND ALFALFA ASH ON STEER PERFORMANCE

(1952 Experiment)[1]

Item	Rations[2]			
	Basal	Basal Plus Reduced Iron	Basal Plus Trace Minerals	Basal Plus Alfalfa Ash
Number of steers	3	3	3	3
Av. initial weight, lb.	406	426	405	409
Av. final weight, lb. (140 days)	593.3	596.0	679.6	674.0
Av. daily gain, lb.	1.34	1.21	1.96	1.89
Av. daily ration, lb.:				
Ground ear corn	8.08	7.46	10.1	10.26
Urea ("262"—Du Pont)	.169	.166	.173	.166
Cerelose	1.44	1.41	1.47	1.41
Mature timothy hay	1.67	1.60	1.83	1.63
Feed per 100 lb. gain, lb., av.:				
Ground ear corn	613.4	612.1	533.1	547.5
Urea ("262"—Du Pont)	14.43	14.8	9.67	9.67
Mature timothy hay	128.4	131.3	95.4	86.4
Cerelose	122.6	125.8	82.2	81.6
Total	878.8	884.0	720.4	725.2

[1] L.S.D. for av. daily gain: 5%, 0.55 lb.; 1%, 0.78 lb. Differences in feed intake and feed efficiency were not significant at 5% level.

[2] Mineral supplement, gm. per day per steer: iron, 0.8; alfalfa ash, 75.0; trace mineral mixture, 1.465 (see text).

None of the mineral supplements significantly influenced feed efficiency but there was a decided trend toward lower feed requirements per unit of gain in the mineral supplemented steers.

Digestion Experiment

After the steers had been on the rations described in table 1 for 99 days in the first experiment (1952), a digestion experiment was carried out using the chromic oxide marker technique. The steers were full fed but no attempt was made to increase the amount of ground ear corn

fed during the actual collection period. The averages of these results are presented in table 3.

All lots of steers were about equally efficient in the apparent digestion of dry matter, crude protein, N.F.E., or crude fiber; thus, despite some variations, there did not appear to be consistent trends toward improved

TABLE 2. THE EFFECT OF COBALT OR TRACE MINERALS ON STEER PERFORMANCE

(1953 Experiment)[1]

Item	Rations		
	Basal	Basal Plus[2] Cobalt	Basal Plus[3] Trace Minerals
Number of steers	4	4	4
Av. initial weight, lb.	370.3	380.5	381.3
Av. final weight, lb. (280 days)	722.3	804.8	872.6
Av. daily gain, lb.	1.26	1.52	1.75
Av. daily ration, lb.:			
Ground ear corn	5.97	7.91	8.95
Supplement, lb.[4]	2.42	2.41	2.44
Mature timothy hay	2.03	2.04	2.03
Feed per 100 lb. gain, lb.:			
Ground ear corn	475.0	522.0	509.0
Mature timothy hay	162.0	134.0	116.0
Supplement	192.0	159.0	139.0
Total	829.0	815.0	764.0

[1] L.S.D. for Av. daily gains: 5%, 0.34 lb.; 1%, 0.48 lb.
L.S.D. for Corn intake: 5%, 1.24 lb.; 1%, 1.75 lb.
[2] 0.5 mg. cobalt per day per steer.
[3] Mixture of manganese, copper, zinc, cobalt and manganese. See text for amounts fed.
[4] Crystalline urea (Du Pont) was used in this study. Supplement mixture composed of urea, cerelose, and minerals.

or reduced efficiency of digestion of the ration as a result of trace minerals, iron, or alfalfa ash supplementation.

As might be expected when the Wilcox-Moxon method for crude fiber was used, the percentage fiber digestion dropped and the utilization of the N.F.E. fraction increased. This can be explained by the shifting of the lignin from the N.F.E. fraction to the crude fiber fraction (Moxon and Bentley, 1953).

As with the A.O.A.C. method, the data obtained with the Wilcox-Moxon method do not show a trend to indicate that mineral feeding affects the apparent digestibility of the ration.

Vitamin B_{12} Analyses

Since a growth response had been obtained by feeding cobalt to the steers and because of the known relationship between cobalt and vitamin B_{12} synthesis in ruminants (Abelson and Darby, 1949; Marston, 1952), it seemed logical to analyze the feces and livers from these animals for vitamin B_{12}. The results of these microbiological analyses

TABLE 3. APPARENT DIGESTIBILITY OF BASAL AND MINERAL SUPPLEMENTED RATIONS

(1952 Experiment)[1]

Item	Basal	Basal Plus Iron	Basal Plus Trace Minerals	Basal Plus Alfalfa Ash
Number of steers	3	3	3	3
Dry matter intake, lb. per day	10.70	10.07	13.18	12.66
Protein, as percent of intake	11.93	12.04	11.36	11.24
Crude fiber, as percent of intake	10.87	10.94	10.67	10.61
Digestion coefficients:				
Dry matter, percent	63.97	66.08	64.37	64.75
Crude protein, percent	52.70	55.45	56.44	51.84
Crude fiber, percent, A.O.A.C.	40.61	34.32	37.39	43.86
Crude fiber, percent, W-M[2]	33.79	29.15	33.79	34.66
N.F.E., percent, A.O.A.C.	69.55	70.81	69.19	69.99
N.F.E., percent, W-M	81.67	85.98	81.62	82.14

[1] The average of the results by dietary treatments. Analyses were on composite samples from an 8 day collection period and the results are reported on a moisture-free, ash-free basis.
[2] Analyses for crude fiber by the Wilcox-Moxon procedure.

of livers, blood plasma, and composite samples of feces are summarized in table 4.

Feeding either alfalfa ash, a trace mineral mixture which contained cobalt, or cobalt alone to steers in both experiments significantly increased the amount of liver vitamin B_{12} 7–10 fold. Likewise, the vitamin B_{12} activity was roughly ten times greater in the feces from steers fed cobalt or a cobalt containing mineral supplement than in feces from steers fed the basal rations.

Mineral Determinations

Early in these studies, analyses were made for the iron, copper, manganese, and cobalt content of the basal ration. The ration had the following cobalt content, p.p.m.: mature timothy hay, 0.06–0.07;

ground ear corn, 0.03–0.06; and the supplement less than 0.01. Dr. K. C. Beeson of the U. S. Plant, Soil, and Nutrition Laboratory, Ithaca, N. Y., kindly analyzed a sample of the poor hay and found 0.06 p.p.m. cobalt. If one calculates the over-all cobalt content of the ration based on the average feed consumption of the steers fed the basal ration, the cobalt content was from 0.03 to 0.04 p.p.m.

TABLE 4. THE EFFECT OF TRACE MINERALS, ALFALFA ASH, AND COBALT ON THE VITAMIN B_{12} CONTENT OF LIVER, DRIED FECES, AND BLOOD PLASMA[1, 2, 3]

	1952 Experiment		1953 Experiment (4)			
		Liver,	Blood	Kidney	Liver	Dried
Ration	Dried Feces μg/gm	Wet Weight μg/gm	Plasma μg/100 ml.	μg/gm, wet		Feces μg/gm
Basal plus Fe, Mn, or Cu	.064 (3)	.066 (3)	0.007	.053	.116	0.243
Basal plus cobalt		.3 (2)	0.018	.21	.68	1.2
Basal plus trace mineral mixture	.863 (3)	.32 (3)	0.014	.22	.69	1.52

[1] Alkali stable vitamin B_{12} activity for *L. leichmannii* in liver and kidney was 0.003 and 0.018 μg/gm, respectively, which was relatively constant for all samples.

[2] The results are averages and the numbers in parentheses indicates the number of steers sampled.

[3] L.S.D. for Liver: 1%, 0.37; 5%, 0.268.
L.S.D. for Kidney: 1%, 0.12; 5%, 0.08.
L.S.D. for Blood plasma: 1%, 0.008; 5%, 0.006.
Statistical analysis was not applied in the case of dried feces as the results obtained were on composite samples.

The level of cobalt in the liver reflected the change in cobalt intake, as indicated by the following results (p.p.m. cobalt): basal steers (1952), 0.018; (1953), 0.01; basal plus trace minerals (1952), 0.045; basal plus cobalt, 0.03 (L.S.D., 1% equal to 0.013 p.p.m.). All cobalt analyses are expressed on a dry weight basis.

Samples of the poor hay were found to contain 44–63 p.p.m. manganese, 5.5 to 7.7 p.p.m. copper, and 120–320 p.p.m. iron. The corn and cob meal contained 7.6 p.p.m. manganese, 3.2 p.p.m. copper, and 280 p.p.m. iron.

Blood Studies

Samples of blood were obtained from steers in both experiments. Determinations for hemoglobin, blood cell volume (hematocrit), plasma protein, total protein, inorganic phosphorus, ammonia, and urea gave values which were within the normal range for all steer calves irrespective of the rations being fed. It seems pertinent that blood values were not abnormal for the nitrogenous constituents studied despite the fact that the steers were receiving a rather high level of urea in their rations, i.e. about 40 percent of their total nitrogen intake.

Discussion

Steers consuming a limited amount of hay low in minerals, and a relatively greater amount of ground ear corn apparently develop a trace mineral deficiency. This deficiency develops when either soybean oil meal or urea is used as the source of supplementary nitrogen. Besides a trace mineral mixture which contained cobalt, copper, manganese, zinc, and iron, the ashed residues of alfalfa hay or molasses fermentation solubles, molasses, corn silage, or alfalfa meal prevented this deficiency (Klosterman *et al.* 1953).

The trace mineral response obtained in these experiments appeared to be largely attributable to the effect of supplementary cobalt contained in the various mineral supplements used, as judged by growth and feed consumption. Additional evidence for a cobalt effect is indicated by the increase in vitamin B_{12} concentration in the livers, blood, and feces of the steers fed cobalt or cobalt-containing supplements.

Furthermore, cobalt analyses indicate that the cobalt content of the ration was from 0.03–0.04 p.p.m., which is below the level recognized by several workers as being the critical minimum for cattle (Beeson, 1950). It must be pointed out here, that although the hay contained 0.06 p.p.m. of cobalt, the steers were eating only about 1.7–2.0 lb. daily. In addition, all the cobalt in the hay may not have been available to the steers just as the protein and cellulose of a mature hay may be less available than in good quality, less mature roughages.

Cobalt deficiencies in sheep have been reported by several workers in the United States and Australia. The characteristic symptoms in these deficiencies were lack of appetite, low blood hemoglobin, unthriftiness, and loss of weight. The cobalt requirement for sheep is recognized as being higher than for cattle. Perhaps in our experiments we did not reach a critical cobalt level which may explain the failure to observe a drop in blood hemoglobin level.

It would appear that the most significant result of the digestion experiment reported in table 3 is that the *rate* of digestion in these steers was enhanced by mineral feeding—that is, the same *percentage* apparent digestibility was maintained by all steers although those receiving trace minerals or alfalfa ash consumed 24 percent more dry matter. Results of a similar nature have been obtained with sheep fed cobalt-deficient rations (Becker and Smith, 1951; Keener *et al.* 1950).

In terms of pounds of TDN over maintenance requirements, it can be shown that because of increased feed intake the "trace mineral" or the "alfalfa ash" steers had about 47 percent more pounds TDN for

growth and fattening than did the remaining 6 steers. The maintenance requirement was calculated on the basis of 0.8 lb. of TDN per 100 lb. body weight (NRC, 1950) using the average actual steer weights of 555 lb. for the "basal" and "basal plus iron" steers and 615 lb. for the "trace mineral" and "alfalfa ash" steers. Although increased daily TDN intake may offer an explanation of improved steer performance, an even more fundamental question is, why the increased appetite? Clark and Quin (1951) reported that molasses and nitrogen additions to poor quality grass hay for sheep accelerated the rate of digestion while the percentage digested was unchanged.

In these experiments cobalt was shown to have a marked effect on the synthesis of vitamin B_{12} by the rumen microflora. Increased vitamin B_{12} synthesis provides the vitamin needs to the host, i.e. the steer which may directly influence the well-being of the animal and indirectly influence appetite. Still another possibility exists—that the bacterial metabolic processes during which vitamin B_{12} is synthesized is important to the maintenance of good ruminal activity. It has been reported that large doses of injected vitamin B_{12} will alleviate a cobalt deficiency in sheep (Becker *et al.*, 1949, 1951; Smith *et al.*, 1951; Marston, 1952) which would suggest that the entire cobalt deficiency syndrome is associated with a lack of vitamin B_{12}.

Summary

In each of two feeding experiments, steer calves were individually fed a ration of mature timothy hay, ground ear corn, urea, cerelose, calcium, phosphorus, iodized salt, and vitamin A. A trace mineral supplement (cobalt, manganese, zinc, iron, and copper) or alfalfa ash significantly increased the average daily gains of the steers. Studies to determine which one of the trace minerals was responsible for the growth stimulation indicated that cobalt was the first limiting trace mineral deficiency although the possibility that other minerals, e.g. manganese, may be beneficial is not excluded.

Besides improved gains, feeding cobalt or a cobalt-containing supplement significantly increased the cobalt and vitamin B_{12} content of the liver, and the amount of vitamin B_{12} excreted in the feces. Based on ration analyses, it was calculated that the average cobalt content of the ration was 0.03–0.04 p.p.m.

Feed intake, chiefly ground ear corn, was significantly increased (one experiment) and there was a trend toward more efficient use of feed by the trace mineral, alfalfa ash, or cobalt-fed steers but the differences

were not significant. The results of one digestion trial using the chromic oxide marker technique indicated that the apparent digestibility of the ration was unaffected by the mineral supplements.

Possible explanations for these findings are discussed.

Literature Cited

Abelson, P. H. and H. H. Darby. 1949. The synthesis of vitamin B_{12} in the digestive system of the sheep. Science 110:566.

Arthur, D., I. Motzok and H. D. Branion. 1953. The determination of cobalt in forage crops. Canadian Jour. Agr. Sci. 33:1.

Association of Official Agricultural Chemists. 1950. "Official Methods of Analysis." George Banta Pub. Co., Menasha, Wis.

Becker, D. E., S. E. Smith and J. K. Loosli. 1949. Vitamin B_{12} and cobalt deficiency in sheep. Science 110:71.

Becker, D. E. and S. E. Smith. 1951. The metabolism of cobalt in lambs. Jour. Nutr. 43:87.

Beeson, K. C. 1950. Cobalt: Occurrence in soils and forages in relation to a nutritional disorder in ruminants—A review of literature. Agric. Info. Bul. No. 7. U.S. Dept. of Agr., Washington.

Bentley, O. G., T. V. Hershberger and A. L. Moxon. 1953. Vitamin B_{12} and cobalt in cattle nutrition. Fed. Proc. 12:177.

Bentley, O. G. and A. L. Moxon. 1952. A semi-synthetic diet for studying the trace element requirements of cattle. JOURNAL OF ANIMAL SCIENCE 11:756.

Bentley, O. G. and P. H. Phillips. 1951. The effect of low manganese rations upon dairy cattle. Jour. Dairy Sci. 34:396.

Bohstedt, G. 1942. Minerals in dairy cattle nutrition: A review. Jour. Dairy Sci. 35:441.

Bolin, D. W., R. P. King and E. W. Klosterman. 1952. A simplified method for the determination of chromic oxide when used as an index substance. Science 116:634.

Burroughs, W. E. 1952. Personal communication.

Burroughs, W. E., P. Gerlaugh and R. M. Bethke. 1950. The influence of alfalfa ash and fractions of alfalfa hay upon the digestion of ground corn cobs. JOURNAL OF ANIMAL SCIENCE 9:207.

Burroughs, W. E., A. Latona, P. DePaul, P. Gerlaugh and R. M. Bethke. 1951. Mineral influences upon urea utilization and cellulose digestion by rumen microorganisms using the artificial rumen technique. JOURNAL OF ANIMAL SCIENCE 10:693.

Clark, R. and J. I. Quin. 1951. Studies on the alimentary tract of the Merino sheep in South Africa XXIII. The effect of supplementing poor quality grass hay with molasses and nitrogenous salts. Onderstepoort Jour. of Vet. Res. 25:93.

Kane, E. A., W. C. Jacobson and L. A. Moore. 1952. Diurnal variation in the excretion of chromium oxide and lignin. Jour. Nutr. 47:263.

Kane, E. A., W. C. Jacobson and L. A. Moore. 1950. A comparison of techniques used in digestibility studies with dairy cattle. Jour. Nutr. 41:583.

Keener, H. A., G. P. Percival, G. H. Ellis and K. C. Beeson. 1950. A study of the function of cobalt in the nutrition of sheep. JOURNAL OF ANIMAL SCIENCE 9:404.

Kitzes, G., C. A. Elvehjem and H. A. Schuette. 1944. The determination of blood plasma iron. Jour. Biol. Chem. 155:653.

Klosterman, E. W., L. E. Kunkle, O. G. Bentley and W. Burroughs. 1953. Supplements to poor quality hay for fattening cattle. Res. Bul. 732. Ohio Agr. Exp. Sta.

Marston, H. R. 1952. Cobalt, copper, and molybdenum in the nutrition of animals and plants. Physiol. Rev. 32:66.

McNaught, M. L., E. C. Owen and J. A. B. Smith. 1950. The utilization of non-protein nitrogen in the bovine rumen. 6. The effect of metals on the activity of the rumen bacteria. Biochem. Jour. 46:36.

Mitchell, H. H. 1947. The mineral requirements of farm animals. JOURNAL OF ANIMAL SCIENCE 6:365.

Moxon, A. L. and O. G. Bentley. 1953. Some discrepancies in A.O.A.C. crude fiber and N.F.E. values in roughages. JOURNAL OF ANIMAL SCIENCE 12:925.

National Research Council. 1950. Recommended nutrient allowances for beef cattle. 2101 Constitution Ave., Washington, D. C.

Parks, R. Q., S. L. Hood, C. Hurwitz and G. H. Ellis. 1943. Quantitative chemical microdeterminations of twelve elements in plant tissue. Ind. and Eng. Chem., Anal. Ed. 15:527.

Russell, F. C. 1944. Minerals in pasture: deficiencies and excesses in relation to animal health. Tech. Comm. No. 15. Imperial Bureau of Animal Nutrition, Rowett Inst. Bucksburn, Aberdeen, Scotland.

Schürck, A. F., L. E. Lloyd and E. W. Crampton. 1950. The use of chromic oxide as an index for determining the digestibility of a diet. Jour. Nutr. 41:629.

Skeggs, H. R., H. M. Nepple, K. A. Valentik, J. W. Huff and L. D. Wright. 1950. Observations on the use of *Lactobacillus leichmannii* 4797 in the microbiological assay of vitamin B_{12}. Jour. of Biol. Chem. 184:211.

Smith, S. E., B. A. Koch and K. L. Turk. 1951. The response of cobalt-deficient lambs to liver extract and vitamin B_{12}. Jour. Nutr. 44:455.

Swift, R. W., R. L. Cowan, G. P. Barron, K. H. Maddy and E. C. Grose. 1951. The effect of alfalfa ash upon roughage digestion in sheep. JOURNAL OF ANIMAL SCIENCE 10:434.

Thompson, H. T., L. S. Dietrich and C. A. Elvehjem. 1950. The use of *Lactobacillus leichmannii* in the estimation of vitamin B_{12} activity. Jour. Biol. Chem. 184:175.

Tillman, A. O., C. F. Chappel, R. J. Sirny and R. MacVicar. 1954. The effect of alfalfa ash upon the digestibility of prairie hay by sheep. JOURNAL OF ANIMAL SCIENCE 13:417.

Wilcox, R. and A. L. Moxon. 1949. An improved method for the determination of crude fiber and nitrogen free extract in feeds. Proc. S. Dak. Acad. Sci. 28:51.

THE EFFECT OF VITAMIN A STORES AND CAROTENE INTAKE OF BEEF COWS ON THE VITAMIN A CONTENT OF THE LIVER AND PLASMA OF THEIR CALVES [1]

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In studies of the vitamin A and carotene nutrition of beef cattle, Guilbert and Hart (1935) observed that cows maintained on a low carotene intake were unable to secrete sufficient vitamin A in the milk to protect their nursing calves against avitaminosis A. These results were confirmed and extended by Baker *et al.*, 1953. The maternal liver stores of vitamin A in both these experiments were low, and hence it was uncertain whether the inadequate nutrition of the calf was due to maternal carotene deprivation, low body reserves, or both.

The experiment reported herein was designed to determine whether cows with high initial liver stores of vitamin A could transfer sufficient vitamin A to their milk to protect their calves against a deficiency under conditions of carotene deprivation.

Experimental Procedure

In November, 1952, twenty two-year-old Hereford cows were selected. All had calved in the range herd at approximately 2 years of age and had nursed calves on native grass pastures during the preceding summer, and all were in the fourth and fifth months of pregnancy. They were divided into four lots of five cows each on the basis of body weight, previous calf production, and expected calving date. The experimental treatments of the cows in each lot were: lot I, no supplemental carotene; lot II, supplemental carotene only during lactation; lot III, supplemental carotene only during gestation; and lot IV, supplemental carotene during both gestation and lactation. The basal ration fed each cow during gestation was cottonseed meal, 2½ lb.; ground milo, 1 lb.; and wheat straw *ad libitum*. During lactation, the daily cottonseed meal allowance was increased to three lb. and the ground milo to four lb. Salt and a mineral mixture of equal parts of ground limestone, steamed bone meal, and salt were available to the cows at all times during gestation and lactation.

[1] Published with the approval of the Director, Oklahoma Agricultural Experiment Station.
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A crude carotene concentrate[3] derived from alfalfa and containing approximately 6 mg. of carotene per gram was used as a source of supplemental carotene. The carotene concentrate was mixed with the grain ration in amounts necessary to provide 60 mg. of carotene per head daily during gestation and 300 mg. during lactation.

Liver biopsy samples were collected by the technique of Whitehair *et al.* (1952) from the cows at the beginning of the experiment, at parturition, and three months postpartum. The calves were biopsied at birth and at three months of age. Blood samples were collected from the cows at monthly intervals before parturition. Additional blood samples were taken from the cows and their calves immediately after parturition and at one week, two weeks, one month, two months and three months postpartum. Colostrum samples were collected immediately after parturition and milk samples were obtained at 7, 14 and 21 days and at the end of 1, 2 and 3 months of lactation. The calves were separated from the cows four hours before milk samples were collected. One quarter of the cow's udder was milked completely to obtain a representative milk sample. Blood, liver, and milk samples were analyzed for carotene and vitamin A. Liver samples were analyzed according to the method of Gallup and Hoefer (1946), plasma by the method of Kimble (1939), and milk by the method of Lesher *et al.* (1945).

Results and Discussion

The average liver levels of vitamin A and carotene for the four lots of cows and their calves are presented in table 1. A summary of the plasma and milk levels of vitamin A and carotene for the cows and of plasma vitamin A for the calves is shown in table 2.

Initial liver stores of vitamin A and carotene of the cows were high, reflecting high carotene intake during the summer grazing season. That vitamin A liver reserves of cows grazing green pasture may be quite variable despite similar treatment was indicated by a range of from 62 to 462 mcg. of vitamin A per gram of dry matter in samples collected in November. During gestation, the liver stores of all cows decreased rapidly with one exception, this being a cow which had less than average liver reserves and to which dietary carotene was fed. Supplementation of the rations of the cows of Lots III and IV with 60 mcg. of carotene per head daily did not materially retard the rate of depletion of the liver stores of vitamin A. However, the addition of 300 mg. to the daily

[3] Generously supplied by the Chlorophyll Chemical Corporation, McAllen, Texas.

ration during lactation (Lots II and IV) did tend to spare liver stores of vitamin A and actually increased the liver levels of carotene. Cows that received no supplemental carotene during lactation (Lots I and III) showed a continued and accelerated decrease in vitamin A liver stores during lactation.

TABLE 1. AVERAGE VITAMIN A AND CAROTENE CONTENT OF LIVERS OF COWS AND THEIR CALVES

(Mcg./gm. Dry Matter)

Liver Component	Lot No.	5 mo. Prepartum	At Parturition	3 mo. Post Partum
		Cows		
Vitamin A	I	302.9±122.8	141.3± 76.8	31.4± 29.3
	II	307.6±105.7	138.4±105.6	111.4± 61.4
	III	151.2± 72.5	56.3± 18.8	9.7± 9.9
	IV	251.7±149.8	112.3± 58.5	84.0± 48.5
Carotene	I	20.2± 5.6	4.7± 3.1	2.5± 2.3
	II	18.7± 4.0	5.3± 1.7	11.1± 3.3
	III	11.6± 3.7	5.7± 2.0	1.8± 1.1
	IV	17.3± 3.3	6.9± 1.7	9.8± 3.8
		Calves		
Vitamin A	I		3.9± 1.9	3.3± 1.9
	II		5.7± 3.0	15.9± 7.1
	III		2.7± 1.1	1.7± 0.6
	IV		2.9± 1.1	13.3± 7.0

Liver stores of vitamin A and carotene of the newborn calves did not appear to be related to treatment of the cows during gestation or to their liver stores at parturition. These results are in agreement with those previously observed (Baker *et al.*, 1953), and with those reported by Thomas *et al.* (1947), and Guilbert and Hart (1935). It is apparent from the reports of Braun and Carle (1943), and Eaton *et al.* (1949) that extremely high intakes of carotene or vitamin A are necessary before substantial placental transfer occurs. Supplementation of the rations of the cows during lactation (Lots II and IV vs. Lots I and III) produced a rather marked increase in the calves' liver stores of vitamin A at three months of age. The plasma vitamin A levels of the calves (table 2) from one to three months of age corresponded to the carotene intake of their dams during lactation, whereas during the first two weeks no such effect was observed.

TABLE 2. AVERAGE VITAMIN A AND CAROTENE CONTENT OF PLASMA AND MILK OF COWS AND VITAMIN A CONTENT OF PLASMA OF THEIR CALVES

(Mcg./100 ml.)

Item	Lot No.	5 mo. Prepartum	1 mo. Prepartum	At Parturition	Lactation 1 wk.	2 wk.	1 mo.	2 mo.	3 mo.
				Cows					
Plasma vitamin A	I	32.5	25.0	17.3	14.4	17.0	17.2	18.6	15.5
	II	30.0	21.1	12.2	14.3	17.9	15.6	15.4	26.5
	III	27.4	20.9	13.4	12.2	12.6	12.5	12.1	16.9
	IV	34.0	25.5	16.8	12.3	17.1	19.6	22.8	30.4
Plasma carotene	I	111.6	22.9	21.3	19.8	21.2	20.8	18.4	18.6
	II	102.9	21.6	17.5	89.4	127.5	147.9	149.1	151.7
	III	73.8	41.4	48.1	28.1	26.9	23.3	17.2	19.5
	IV	97.5	49.1	63.0	89.6	113.2	101.1	117.0	92.3
Milk and colostrum Vitamin A	I			144.0	6.2	5.3	6.2	6.3	3.8
	II			224.5	11.0	7.8	9.0	8.8	8.6
	III			128.3	6.5	5.7	3.8	3.7	3.3
	IV			133.2	10.0	7.8	7.6	6.6	7.2
				Calves					
Plasma vitamin A	I			4.9	13.2	11.6	7.9	6.0	4.5
	II			3.8	12.0	11.5	9.5	9.0	12.6
	III			6.2	8.6	7.3	5.9	4.6	5.6
	IV			4.9	13.6	13.8	10.3	10.0	11.5

The plasma carotene levels of the cows at the beginning of the experiment were indicative of the previous high carotene intake. The effect of carotene supplementation on the plasma carotene levels was not apparent until the third month of the experimental period. During lactation the plasma carotene levels of the cows receiving supplemental carotene were comparable to those reported by Long *et al.* (1950) for cows receiving a good quality roughage. Plasma vitamin A levels of the cows were essentially normal in all lots until the latter stages of the lactation phase. By the end of the third month of lactation, the cows not receiving supplemental carotene had lower plasma vitamin A levels than those receiving additional carotene. The normal plasma vitamin A levels of the cows of Lots II and IV, coupled with a decreased rate of depletion of liver stores and normal reproduction and lactation performance, suggests that 300 mg. of carotene per head daily is more than adequate to meet the carotene requirements during lactation.

The vitamin A content of the colostrum of the cows of this study was considerably higher than that previously observed at this station (Baker *et al.*, 1953). It is suggested that the higher liver stores of the cows used in this study may have been responsible for this condition. No correlation between the vitamin A reserves of the cow and the vitamin A content of the colostrum was apparent in this study. Milk levels of vitamin A, on the contrary, were related in a positive fashion to carotene supplementation during lactation.

Typical symptoms of avitaminosis A (Guilbert and Hart, 1935) were observed in three calves of Lot III and two calves of Lot I. No symptoms indicative of a vitamin A deficiency were observed during this experiment in any of the cows, regardless of treatment, nor in the calves of Lots II and IV. Thus even cows with ample stores early in gestation appear unable to provide enough vitamin A in milk during lactation to prevent deficiency symptoms in the calves. The practical implications for early calving or fall calving on native ranges in the Southwest are obvious.

Summary

Mobilization of stored vitamin A and carotene by lactating beef cows on a low plane of carotene intake was inadequate to provide sufficient vitamin A for their calves. Vitamin A content of milk, and to a lesser degree carotene, seem much more closely related to dietary carotene intake during lactation than to differential liver stores resulting from various levels of carotene intake prepartum.

Supplementation of low carotene rations with 300 mg. of carotene per cow daily during lactation resulted in normal carotene and vitamin A levels in the plasma and livers of suckling calves. The rate of depletion of the liver stores of the cow during 5 months postpartum was also decreased.

Literature Cited

Baker, F. H., Robert MacVicar and L. S. Pope. 1953. Placental and mammary transfer of vitamin A and carotene by beef cows. Proc. Soc. Exp. Biol. and Med. 83:571.

Braun, W. and B. N. Carle. 1943. The effect of diet on the vitamin A content of the bovine fetal liver. Jour. Nutr. 26:549.

Eaton, H. D., J. A. Christian, F. C. Daugherty, A. A. Spielman, and L. D. Matterson. 1949. The effect of crude soybean lecithin on the absorption and utilization of vitamin A fed preparatum to the ewe and sow. JOURNAL OF ANIMAL SCIENCE 8:224.

Gallup, W. D. and J. A. Hoefer. 1946. Determination of vitamin A in liver. Ind. Eng. Chem. Anal. Ed. 18:288.

Guilbert, H. R. and G. H. Hart. 1935. Minimum vitamin A requirements with particular reference to cattle. Jour. Nutr. 10:409.

Kimble, M. S. 1939. The photocolorimetric determination of vitamin A and carotene in human plasma. Jour. Lab. Clin. Med. 24:1055.

Lesher, M., J. K. Brody, H. H. Williams and I. G. Macy. 1945. Human milk studies XXVI. The vitamin A and carotene contents of colostrum and mature human milk. Amer. Jour. Dis. Child. 70:182.

Long, R. A., W. J. Van Arsdell, Robert MacVicar and O. B. Ross. 1952. Blood composition of normal beef cattle. Okla. Agr. Expt. Sta. Bul. No. T-43.

Whitehair, C. K., D. R. Peterson, W. J. Van Arsdell and O. O. Thomas. 1952. A liver biopsy technique for cattle. Jour. Amer. Vet. Med. Assn. CXXI:285.

USE OF BEEF CATTLE FEEDING DATA IN EVALUATING MOUNTAIN MEADOW MANAGEMENT PRACTICES [1]

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AN interpretation of factors affecting the production of forage crops is more complete and realistic if results can be expressed in terms of animal performance. Usually, yields of dry hay have been taken by the rancher as the final basis for evaluating factors affecting forage production. In certain instances, additional information is available on such quality factors as the percentages of crude protein, phosphorus, or other chemical constituents of the forage. These criteria have not proven entirely satisfactory.

In Gunnison County, Colorado, a test feeding plant for beef cattle was established where the relative merits of various management practices utilized in hay production and feed supplementation could be determined. This report covers a two-year period, 1950–1951, of investigation in which hays produced under variable fertility and harvest management practices were fed to cattle and the results evaluated in terms of beef production.

Procedure

Hay Production

Approximately 60 acres of a fairly representative high altitude meadow were used for the production of hay for subsequent beef cattle feeding experiments. Four fertilizer treatments, i.e., control, nitrogen, phosphorus, and a combination of nitrogen and phosphorus, were applied to separate duplicated portions of the field.

Portions of the hay from each fertilizer treatment were harvested about August 2 (Mid-season cut practice) and approximately September 4 (Late cut practice). In each case, the hay was baled and stored

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for feeding. At the time of feeding, daily samples were collected for chemical determination of nitrogen and phosphorus in the forage. Crude protein percentages were based on total nitrogen multiplied by the factor 6.25.

In conjunction with this feeding study, the relationship between crude protein percentage in hay, which is indicative of quality, and harvest date was studied. This was accomplished by taking hay samples from a large number of ranches in Colorado progressively throughout the season and analyzing them for crude protein content.

TABLE 1. FERTILIZER AND HARVEST MANAGEMENT TREATMENTS EMPLOYED IN THE PRODUCTION OF HAY FED TO LOTS OF BEEF CATTLE IN 1950 AND 1951

Fertilizer Treatment[1] lbs. per Acre N-P	Time of Hay Harvest	Protein Supplement Fed with Hay [2]	Years in which Hay was Fed
0–0	Mid-season	None	1950 and 1951
0–0	Late	None	1950 and 1951
100–0	Mid-season	None	1950 only
0–87	Mid-season	None	1950 and 1951
0–87	Late	None	1951 only
100–87	Mid-season	None	1950 and 1951
100–87	Late	None	1950 and 1951
0–0	Mid-season	43%	1950 only
0–0	Late	43%	1950 only
100–0	Mid-season	39%	1951 only
100–0	Mid-season	20%	1951 only

[1] Nitrogen and phosphorus applied in spring of 1950; nitrogen only was applied in spring of 1951.

[2] Protein supplement was fed at the rate of one pound per day for each animal.

Feeding Experiments

In each of the two years, the test included eight lots of heifers. There were 10 animals to each lot with an initial average weight of approximately 450 pounds. A description of the type of hay and supplemental feed given each lot of cattle in the two years is given in table 1.

The amount of hay fed was approximately 5% in excess of the total daily consumption. Salt and warm water were kept before the cattle at all times. The feeding periods were 120 days starting about the middle of December and ending about the middle of April.

At the outset and at the end of the experiment individual heifers were weighed on two consecutive days. The weight of each pen of cattle and the amount of hay consumed by each lot were determined at two-week intervals.

The averages of daily gain, daily hay consumption and daily crude protein intake for the 120-day feeding periods were computed. Daily crude protein intakes were related to beef gains. This relationship was used to convert forage production data from the Blackstock Mountain Meadow Factorial Experiment at Iola, Colorado, into pounds of beef per acre.

Method of Estimating Beef Production from Protein Yield Data

Estimation of the amount of beef produced from an acre of mountain meadow hay involves the following assumptions:

1. Daily consumption of hay per animal is relatively constant, regardless of the quality of hay fed. This assumption is supported by the data secured during the two years of feeding (table 2). The daily consumption of hay per animal was approximately 10.7 lbs. in 1950 and 12.5 lbs. in 1951.
2. The hay produced on the area in question is similar to hay fed to the test animals so that the regression equations are applicable.

An estimate of acre-production of beef may be calculated by the following steps:

1. Daily consumption of hay per animal multiplied by percent crude protein in hay of any given treatment equals pounds of crude protein consumed by each animal per day.
2. The regression equations (figure 2) are used to calculate pounds daily gain from corresponding available crude protein.
3. Pounds crude protein available per animal per day divided by pounds daily gain per animal equals pounds crude protein required to produce one pound of beef.
4. Pounds hay per acre for any given treatment multiplied by percent crude protein equals pounds crude protein per acre.
5. Pounds crude protein per acre divided by pounds crude protein required to produce one pound of beef equals pounds of beef produced per acre for any given treatment.

Results and Discussion

Chemical analyses of mountain meadow hay samples from several areas show a decline in crude protein from approximately 12 to 6 percent in both 1950 and 1951 as the hays matured and the season progressed from July 10 to September 4 (figure 1). It will be noted from the regression equations that each week's delay in harvest after July 10 caused a reduction of approximately 0.6 percent crude protein in the hay.

The data from the feeding tests show that the daily hay consumption was lower in 1950 than in 1951, 10.7 lbs. per day for each animal compared to 12.5 lbs. As a result of the increased hay consumption, more

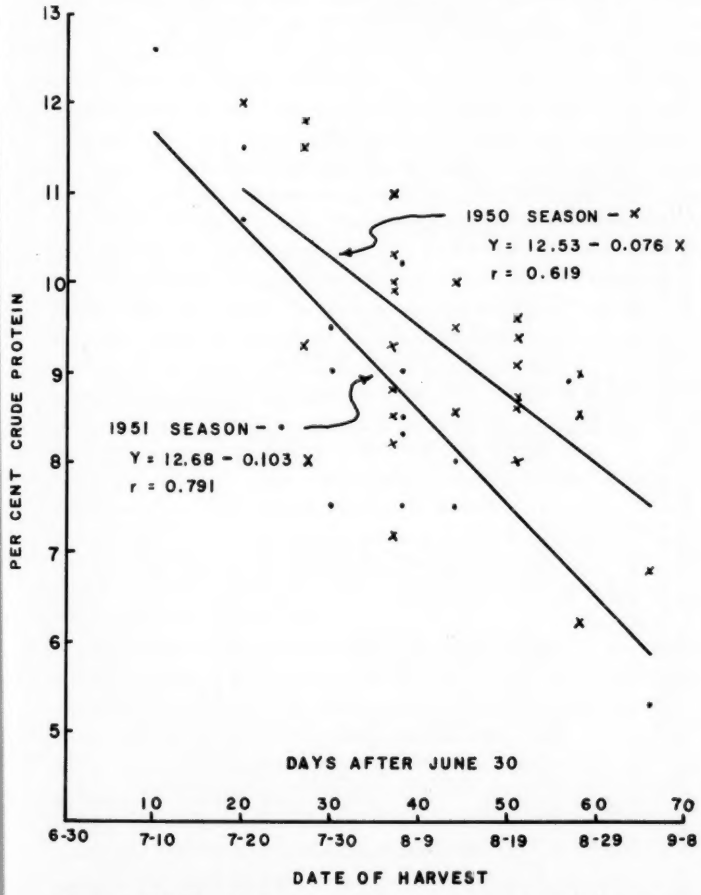

Figure 1. Relationship between per cent crude protein and date of harvest of Colorado mountain hays, 1950–1951 (data from several Colorado ranches).

crude protein was consumed from the mid-season cut in 1951 than in 1950. However, the 1951 late cut hays were lower in protein content than in 1950 (table 2) and increased hay consumption was not sufficient to offset the decline in quality. As a result, the crude protein intake

TABLE 2. FEEDING VALUE OF HIGH ALTITUDE MEADOW HAYS AS DETERMINED USING A GROUP OF 450-POUND HEIFER CALVES. GUNNISON, COLORADO, 1950–1951.

Designation of Rations		Lb. Hay Consumed Daily		Lb. Hay per. lb. Gain		% Crude Protein in Hay		Crude Protein Available Daily-Lb.		Daily Gain Lb.	
Hay	Supplement	1950	1951	1950	1951	1950	1951	1950	1951	1950	1951
OM	S_1	11.2		7.9		9.2		1.452[1]		1.40	
OL	S_1	10.6		9.3		7.0		1.168[1]		1.14	
OM	..	10.8	12.7	10.5	15.8	8.0	8.4	.863	1.059	1.03	.80
OL	..	10.6	12.2	13.5	101.1	7.0	5.4	.748	.658	.79	.12
NPM	..	10.4	12.7	9.7	15.2	9.7	9.1	1.006	1.157	1.08	.84
NPL	..	10.7	12.3	13.7	60.1	6.7	5.3	.711	.647	.78	.24
NM	..	10.9		9.9		9.5		1.041		1.10	
PM	..	10.6	12.3	9.5	14.2	8.8	8.8	.935	1.082	1.12	.87
OM	S_2		12.8		14.1		8.7		1.311[1]		.91
OM	S_3		12.9		11.8		9.0		1.547[1]		1.09
PL	..		12.3		60.1		5.2		.634		.20

Key: *Hays from the following treatments:*
O=No soil treatment
N=Soil treatment 100 lbs. Nitrogen per acre/year
P=Soil treatment 200 lbs. P_2O_5 per acre initially
M=Mid-season cut hay (about August 2)
L=Late cut hay (September 4)

Protein supplementations:
S_1=1 lb. 43% per day
S_2=1 lb. 20% per day
S_3=1 lb. 39% per day

[1] Includes protein from supplement.

was less in 1951 than in 1950 from late cut hays. The stage of hay maturity apparently did not affect the amount of hay consumed in either year (table 2).

It appears that the cattle were unable to utilize the hay as efficiently in 1951 as in 1950. Table 2 shows that an average of approximately 10 pounds of mid-season cut hay were required per pound of gain in 1950, where 15 pounds were needed in 1951. With late cut hays, the amounts required were approximately 14 pounds in 1950, compared with 71 pounds in 1951. These differences in efficiency are undoubtedly due to many uncontrolled and unmeasured factors, among which might be listed the utilization of crude protein by the animals and the extreme severity of the 1951 winter feeding season which had a mean temperature of 10° F. as compared with 24° F. for the 1950 winter season.

In a comparison between hays produced without fertilizer, cut mid-season and cut late, daily gains were 1.03 versus .79 pounds in 1950 and .80 versus .12 pound in 1951, respectively (table 2).

In both 1950 and 1951 there was a small increase in the rate of gain when the cattle were fed hay produced on soil that had been fertilized with nitrogen or phosphorus or these elements in combination. These increases were not significant statistically in either year.

During the two years, daily intake of protein per animal ranged from .634 to 1.547 pounds. Within this range the daily rate of gain varied from .12 to 1.40 pounds per day (table 2). As shown in figure 2, a direct relationship between daily gain and crude protein available was found for both years. The coefficients of correlation were $r = 0.946$ for 1950 and $r = 0.962$ for 1951.

Linear regression equations were calculated (figure 2) and show that if crude protein is available in amounts up to approximately 1.6 pounds daily the first pound of crude protein consumed has resulted in a daily beef gain of 1.039 pounds in 1950 and only 0.572 pounds in 1951. Each additional pound of crude protein consumed resulted in a gain of 0.808 pound in 1950 and 1.083 in 1951. It is apparent from figure 2 that if the straight line relationship shown in the 1951 curve continues beyond the intersection with the axis at about 0.4 pound protein the gains would be negative.

An example of how agronomic practices can be expressed in terms of pounds of beef per acre, using the above-mentioned regression equations, is shown in table 3. Crude protein yields from various fertilizer, irrigation, and harvest treatments on the Blackstock Ranch at Iola, Colorado, in 1950 and 1951 are converted to pounds of beef

per acre. It was assumed that the daily hay consumption per animal for each year is equal to that reported in table 2, i.e., 10.7 lbs. in 1950 and 12.5 lbs. in 1951. It will be noted that highest production of beef per acre in 1950 resulted from an early cut and aftermath harvest with

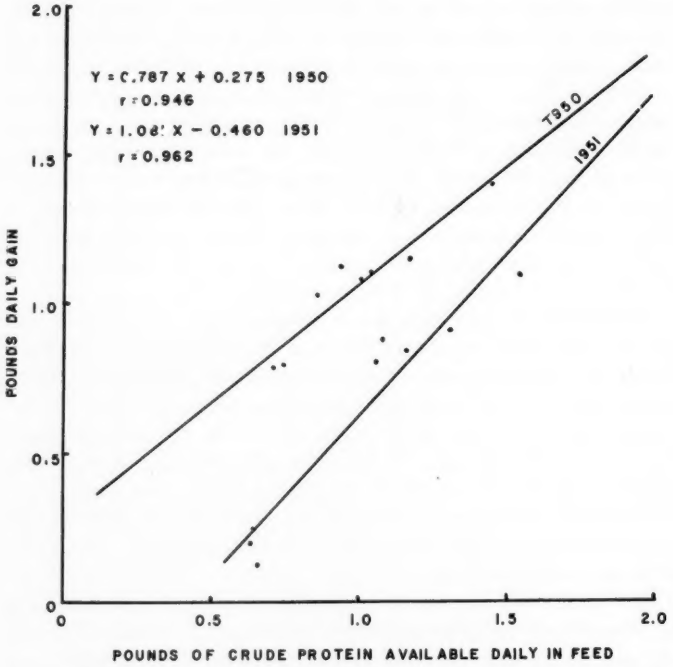

Figure 2. Daily gain (Y) of 450 pound heifers as affected by pounds crude protein available daily.

intermittent irrigation and 160 pounds of nitrogen per acre. In 1951 the same type of irrigation and harvest management showed highest beef production, but nitrogen fertilization had little effect.

An examination of the data presented in table 3 shows the importance of introducing forage quality as a factor in evaluating agronomic practices used in hay production. For example, under intermittent irrigation with no fertilization in 1951, early cut plus aftermath harvest as compared with late cut harvest gave only an 18 percent increase in hay yield whereas the increase in beef production was 84 percent. A second

TABLE 3. CONVERSION OF DATA FROM BLACKSTOCK MOUNTAIN MEADOW HARVEST, IRRIGATION AND FERTILIZATION PRACTICES TO POUNDS BEEF PER ACRE ON A CRUDE PROTEIN BASIS

Iola, Colorado

Practises			Results						Calculated	
		Applied Nitrogen Lb.	Tons Hay per Acre		Percent Crude Protein		Lb. Crude Protein		Lb. Beef per Acre Crude Protein Basis	
Harvest	Irrigation	per Acre	1950	1951	1950	1951	1950	1951	1950	1951
Early cut	Intermittent	0	1.28	2.55	12.03	13.06	309	671	309	537
Aftermath	"	0	0.26	1.16	14.26	18.05	73	424	71	369
Both	"	0	1.54	3.71			382	1095	380	906
Late cut	"	0	1.81	3.14	8.46	10.53	309	664	336	492
Early cut	"	160	2.46	3.18	13.60	11.93	662	754	643	594
Aftermath	"	160	0.41	1.15	12.73	13.72	101	314	100	255
Both	"	160	2.87	4.33			763	1068	743	849
Late cut	"	160	3.45	4.14	8.59	8.66	573	712	623	468
Early cut	Continuous	0	1.09	1.86	11.63	9.19	255	338	258	227
Aftermath	"	0	0.26	0.52	10.52	12.82	56	132	58	106
Both	"	0	1.35	2.38			311	470	316	333
Late cut	"	0	1.75	2.77	7.86	6.85	274	373	308	213
Early cut	"	160	2.49	2.82	12.85	10.23	634	561	628	401
Aftermath	"	160	0.32	0.51	13.27	11.41	85	121	83	92
Both	"	160	2.81	3.33			719	682	711	493
Late cut	"	160	3.35	3.83	8.08	7.13	526	545	578	306

example from the 1951 data shows that with the early cut and aftermath harvest, intermittent irrigation without fertilization as compared with continuous irrigation increased hay yields 56 percent while the increase in beef production was 172 percent. A striking example of the need for introducing quality as a factor in evaluating agronomic practices exists with the 1951 combination of early cut plus aftermath harvest and intermittent irrigation (no fertilization) as compared with late cut harvest and continuous irrigation. The former combination of practices increased hay yield only 34 percent, yet on a beef production basis, the increase was 325 percent.

Summary and Conclusions

Early cut hay is far superior to late cut hay in feeding value. In 1950 one pound of 43% crude protein cake supplement per animal per day was required to raise the feeding value of a late cut hay ration to equal that of early cut hay.

The use of nitrogen and phosphorus fertilizers did not affect the feeding value of the hay significantly.

A direct relationship was found between pounds of crude protein available daily in a ration and the rate of animal gain. A method is suggested for utilizing this relationship in converting yield and protein analysis data into corresponding beef production. The method has possible value in interpreting the results of water control, harvest management, fertility, or other practices in terms of the end product of a ranch —pounds of beef per acre.

Analysis of agronomic data may be made on the dry matter basis; however, where forage is used exclusively for livestock feed, combining dry matter and quality to express the data as livestock gain gives a more complete picture of the results obtained.

THE EFFECT OF AGE OF CASTRATION UPON RATE AND ECONOMY OF GAIN AND CARCASS QUALITY OF BEEF CALVES [1]

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IN the production of beef cattle it is a common and a recommended practice to castrate bull calves at a young age. When a portion of the calves is to be retained for breeding purposes, however, it is difficult to select, at that time, those animals which will make the most valuable sires. When the calves are more fully developed at weaning age a more accurate selection can be made. Recent beef cattle breeding research has shown that post-weaning growth studies are a valuable aid in the selection of beef sires. Information has been needed on the slaughter value of those bulls not retained for breeding purposes.

Experiments conducted at the Arizona Agricultural Experiment Station (1946–1947) showed that fattening bulls made more rapid and economical gains than steers. In one experiment the bulls realized a considerably greater net profit and in a second experiment, a slightly smaller profit than the steers; the bulls produced carcasses of a very satisfactory grade.

The two experiments reported here were conducted to compare the rate and economy of gain in the feed lot and the carcass quality of Hereford calves castrated at one month or at approximately seven months of age to those fed and slaughtered as bulls.

Procedure

The Hereford calves used in these experiments were obtained from a rancher in Nebraska. One-third of the calves were castrated at about one month of age. The other two-thirds were allowed to remain as bulls until weaning at which time they were shipped to the Ohio Agricultural Experiment Station. Upon arrival, a second group of one-third was

[1] Approved for publication as Journal Article N. 32–54.
[2] Deceased.

castrated and the remainder were fed as bulls. The three lots were hand, full fed a standard ration in dry lot. The length of the feeding periods and the average daily rations fed are given in tables 1 and 2.

In the second experiment, the birth and weaning weights of each

TABLE 1. FEED LOT PERFORMANCE OF EARLY AND LATE CASTRATED STEERS AND BULLS

1950–1951 Experiment

Lot		1	2	3
		Steers Castrated May 11, 1950	Steers Castrated Oct. 31, 1950	Bulls
Number in lot		10	10	10
Initial weight, Nov. 7, 1950,	lb.	378.	370.	368.
Final weight, July 17, 1951,	lb.	879.	870.	930.
Av. daily gain, 252 days,	lb.	2.00	2.00	2.23
Av. daily ration:				
Corn and cob meal,	lb.	9.9	9.9	10.4
Soybean oil meal,	lb.	1.5	1.5	1.5
Hay,	lb.	1.4	1.4	1.4
Silage,	lb.	5.0	5.0	5.0
Minerals,	oz.	1.1	1.0	1.1
Salt,	oz.	.5	.7	.5
Feed required per cwt. gain:				
Corn and cob meal,	lb.	495.	500.	467.
Soybean oil meal,	lb.	75.	76.	67.
Hay,	lb.	73.	73.	65.
Silage,	lb.	251.	252.	224.
Minerals,	lb.	3.	3.	3.
Salt,	lb.	2.	2.	1.
Feed cost per cwt. of gain		$16.16	$16.29	$14.97
Appraised selling price per cwt.		$34.02	$34.38	$30.40

Feed Prices Used

Corn and cob meal	—$ 1.50 per 70 lb.	Silage	—$12.00 per ton
Soybean oil meal	— 80.00 per ton	Minerals	— 3.00 per cwt.
Hay	— 25.00 per ton	Salt	— 1.50 per cwt.

individual calf were obtained. Thus, a limited amount of information was secured on the influence of castration upon rate of gain to weaning age.

At the close of the feeding experiments an appraised market value was placed on each animal by experienced cattle buyers. All of the cattle were slaughtered through the Meats Laboratory where detailed slaughter and carcass data were obtained.

Results

In the 1951–1952 experiment, the birth date, birth weight, and weaning weight of each individual calf were obtained. When the weaning weights were adjusted to a standard age of 190 days it was found that the calves which had been castrated weighed an average of 15.8 pounds less than the bull calves. The calves castrated after arrival at Wooster were retarded by a very similar amount so that the initial weights of the two lots of steers were practically identical at the start of the feeding experiment.

The results obtained from the two feeding experiments are presented in tables 1 and 2.

Age of castration, whether shortly after birth or at weaning time, had no influence upon subsequent rate of gain in the feed lot. The rates of gain of early and late castrated steers were identical in the first experiment and nearly so in the second. In both experiments the bulls gained at a significantly faster rate than either lot of steers. In the first experiment the three lots were fed for the same length of time and the bulls gained 0.23 lb. per head daily faster than the steers. In the second experiment, the steers were fed for a longer period in an attempt to reach the same final weight as the bulls. Thus, the steers were fed later into warmer weather and to a higher finish than the bulls and their average daily gain for the entire period averaged 0.48 lb. less than the daily gain of the one lot of bulls.

It will be noted that the bulls consumed slightly more corn and cob meal per head daily than the steers but, because of the faster rate of gain, they required considerably less feed to produce one hundred lb. of gain. The feed cost per cwt. of gain was $1.25 less for the bulls than the average for the steers in the first experiment and $3.78 less in the second experiment.

The appraised selling prices given in tables 1 and 2 are averages of market evaluations placed on each individual animal at time of slaughter. These evaluations were made by regular experienced cattle buyers from the Columbus yards. It will be noted from these data that, if the steers and bulls from these experiments had been sold on the open market, the steers would have sold for approximately $4.00 per cwt. more than the bulls.

After the feeding experiments were completed all animals were trucked, usually in groups of five or six per week, to the Meats Laboratory at Ohio State University, where they were slaughtered. One-half of each carcass was separated into bone, fat trim, and edible por-

tion. The edible portion of the carcass is designated as edible meat with no more than three-eighths inch layer of fat on any surface. Shell-loin steaks, taken from the other half of each carcass, were broiled and

TABLE 2. FEED LOT PERFORMANCE OF EARLY AND LATE CASTRATED STEERS AND BULLS

1951–1952 Experiment

Lot		1	2	3
		Steers Castrated Early	Steers Castrated Nov. 9, 1951	Bulls
Number in lot		12/3*	12/3*	12
Initial weight, Nov. 15, 1951,	lb.	365.0	366.0	384.0
Final weight,	lb.	848.0	844.0	894.0
Average daily gain,	lb.	(247 days) 1.96	(247 days) 1.94	(210 days) 2.43
Average daily ration:				
Corn and cob meal,	lb.	10.5	10.2	10.6
Soybean oil meal,	lb.	1.5	1.5	1.5
Hay,	lb.	2.5	2.5	2.6
Silage,	lb.	5.0	5.0	5.0
Minerals,	oz.	.2	.2	.3
Salt,	oz.	.4	.4	.5
Feed per cwt. of gain:				
Corn and cob meal,	lb.	535.0	529.0	436.0
Soybean oil meal,	lb.	77.0	78.0	62.0
Hay,	lb.	127.0	128.0	108.0
Silage,	lb.	255.0	258.0	206.0
Minerals	lb.	.8	.8	.7
Salt,	lb.	1.0	1.0	1.0
Feed cost per cwt. of gain		$20.68	$20.61	$16.86
Appraised selling price per cwt.		$33.42	$33.35	$29.00

* Three steers from Lots 1 and 2 sent to slaughter on July 3, July 10, July 24, and August 7. Average daily gain calculated on actual steer days on experiment.

Feed Prices Used

Corn and cob meal	$ 1.75 for 70 lb.	Silage	$12.00 per ton
Soybean oil meal	100.00 per ton	Minerals	3.00 per cwt.
Hay	30.00 per ton	Salt	1.50 per cwt.

used to determine palatability and tenderness by a taste panel. The organoleptic tests were patterned after the techniques of Deatherage and Reiman (1946). The results of these slaughter and cut-out experiments are presented in table 3.

In both experiments the bulls were heavier than the steers at time of slaughter. For this reason, the data in table 3 are expressed as percent of live weight or of carcass weight rather than actual weights.

Statistical analyses of the data in table 3 show no significant differences between lots in weight of head as percent of live weight or in amount of bone, expressed as percent of carcass weight. The differences in dressing percentage were significant at the 5 percent level with the bulls having the lower dressing percentage. The following differences

TABLE 3. SLAUGHTER AND CARCASS DATA OF EARLY AND LATE CASTRATED STEERS AND BULLS

Item	1950–1951 Experiment			1951–1952 Experiment		
	Steers Castrated Early	Steers Castrated at Weaning	Bulls	Steers Castrated Early	Steers Castrated at Weaning	Bulls
Number of cattle	10	10	10	12	12	12
Average slaughter weight, lb.	853.9	848.8	916.2	816.8	822.8	848.1
Dressing percentage	60.9	61.2	59.8	59.6	60.1	59.2
Head, percent of live weight	3.13	3.09	3.20	3.12	3.07	3.20
Hide, percent of live weight	8.11	8.48	9.67	8.29	8.08	9.58
Average carcass grade[1]	0.83	1.03	2.23	1.43	1.44	2.78
Forequarter, percent of carcass	52.2	51.5	53.8	51.5	51.5	53.4
Hindquarter, percent of carcass	47.8	48.5	46.2	48.5	48.5	46.6
Kidney knob, percent of carcass	2.50	2.55	1.48	2.30	2.51	1.83
Bone, percent of carcass	15.4	15.6	16.2	16.1	16.2	16.4
Fat, percent of carcass	10.4	10.0	5.4	9.8	9.4	5.7
Edible portion, percent of carcass	73.70	74.10	77.70	74.10	74.40	77.50
Tenderness score[2]	8.10	7.60	7.60	7.60	7.70	6.90
Cost of edible portion per cwt.[3] based on appraised selling price	$75.79	$75.82	$65.43	$75.67	$74.58	$63.21
Av. carcass value per cwt.[4]	$41.22	$40.58	$38.00	$39.89	$39.90	$37.30
Cost of edible portion per cwt. based on av. carcass value[5]	$55.93	$54.76	$48.91	$53.83	$53.63	$48.13

[1] Carcass grade factor—Prime=0, 0.4, 0.7 Choice=1, 1.4, 1.7
Good =2, 2.4, 2.7 Commercial=3, 3.4, 3.7

[2] Tenderness score—10 extremely tender—one extremely tough.

[3] Appraised selling price/dressing percentage/percent edible portion.

[4] Based on wholesale beef carcass prices as follows: Prime, $42.00; Choice, $40.00; Good, $38.00; Commercial, $35.00.

[5] Carcass value/percent edible portion.

were significant at the one percent level. The bulls had heavier hides, a lower carcass grade, heavier forequarters and lighter hindquarters, lighter weight flanks and kidney knobs. The bull carcasses also had less fat trim and a greater percentage of edible portion. There was no significant difference in tenderness rating between the bull and steer carcasses in the 1950–1951 experiment but there was a highly significant difference in the 1951–1952 experiment.

Many of the differences noted between steer and bull carcasses were undoubtedly influenced by the difference in amount of finish present on these carcasses. The carcass grade, dressing percentage, amount of fat trim and percent of edible portion would be directly influenced by the degree of finish of the animals when slaughtered. The steer car-

Figure 1. 1950–1951 Experiment. Lot of early castrated steers. Steer No. 21 from this lot and its carcass.

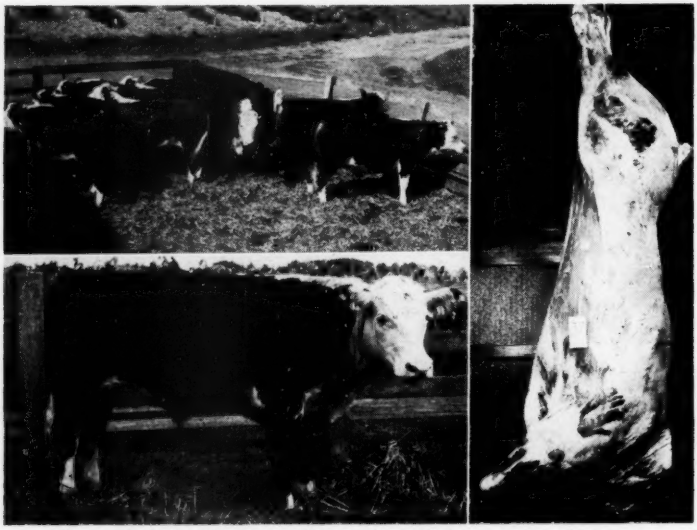

Figure 2. 1950–1951 Experiment. Lot of late castrated steers. Steer No. 37 from this lot and its carcass.

casses had significantly heavier hind quarters but also had significantly heavier kidney knobs than bull carcasses. When the weights of kidney and surrounding fat were subtracted from the hind quarters, analysis of variance showed that the variance due to differences in percent of hind quarter between lots was less than one-half the variance found when the kidney fat was not removed. In addition, the steer carcasses had a larger amount of waste fat in the flank cut than bull carcasses.

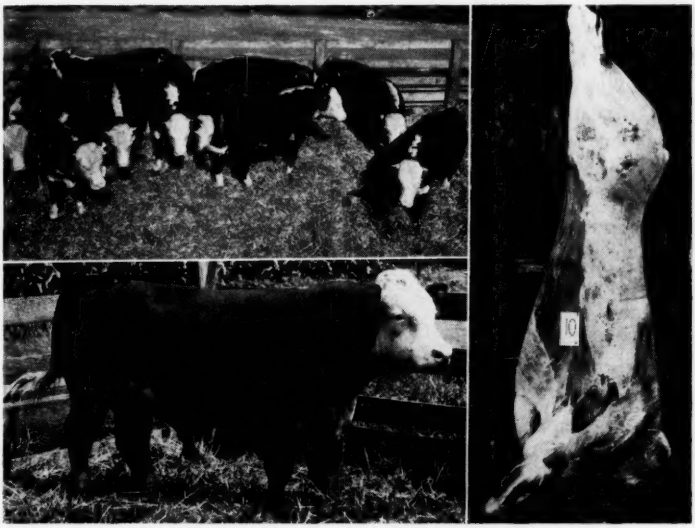

Figure 3. 1950–1951 Experiment. Lot of bulls. Bull No. 10 from this lot and its carcass.

Thus, the proportions of fore and rear quarters were also influenced by the degree of finish. The results of these experiments indicate that there is very little difference between steers and fat, young bulls in proportion of valuable, edible meat in the rear quarter. There was also a highly significant correlation between carcass grade and tenderness score. The carcasses with the higher grade of finish tended to be more tender.

There were no significant differences in any of the slaughter or carcass determinations between steers castrated shortly after birth or those castrated at weaning age.

It will be noted in table 3 that when the cost of edible portion is based on the appraised selling price, the bull carcasses supplied a

pound of edible meat for ten to twelve cents less than the steer carcasses. This difference is somewhat less, five to seven cents, when the cost of edible portion is based on the wholesale value of the carcasses obtained.

Discussion

The results of these two experiments show no significant differences between early and late castrated steers in rate or economy of gain in the feed lot, dressing percentage, or carcass quality. Bull calves were heavier at weaning but their gains were sufficiently retarded immediately following castration that their weight was very similar to the early castrated steers when the two groups were started on feed. Castration at the heavier weights is more difficult and likely to be more hazardous. However, for the breeder who wishes to retain a portion of his male calves for breeding purposes, the advantages of a more accurate selection at weaning time would seem to exceed any disadvantages of late castration.

It is possible that the male hormones produced by normal bulls have a definite stimulus to growth. This was shown by the fact that even though bulls gained significantly faster than steers they were not as fat at time of slaughter. Due largely to a smaller amount of fat trim, the bull carcasses produced a larger proportion of edible meat than the steer carcasses. This difference between castrated and uncastrated males is in agreement with experiments conducted with sheep and swine. Hunt, Meade, and Carmichael (1938) found no significant differences in rate of gain made by rams and wethers but ram carcasses had a higher percentage of lean in the rib cuts than wether carcasses. Bratzler and co-workers (1954) found that boars and late castrated barrows had less backfat and a higher percentage of lean than early castrated barrows.

The beef produced by steer carcasses was slightly more tender than that from bulls. This difference, however, was undoubtedly influenced by the difference in carcass grade between steers and bulls. Beef produced by bull carcasses was of very acceptable quality, no undesirable flavor or aroma was detected. This is in agreement with results from rams (Hunt *et al.*, 1938) but not from boars (Bratzler *et al.*, 1954).

The results of these experiments show that bull calves will make more rapid gains than steer calves when fattened in dry lot. They will also produce more lean meat of comparable quality with less feed per unit of gain.

Due to a greater stimulus for growth, bulls are slower to fatten to the higher grades than steers. In these experiments the bulls and steers were fed in an open shed with relatively small outdoor lots. Under these conditions, very little difference was noted between bulls and steers in amount of restlessness in the feed lot or in handling the animals during feeding and weighing.

Whether or not it would be an economical practice to fatten bulls rather than steers would depend upon the degree of finish desired and the discrimination of the market where the animals were sold. The amount of discrimination would undoubtedly be influenced by the supply and demand for beef of a particular grade.

Summary

Two experiments are reported in which the feed lot performance and carcass quality of early and late castrated steers and bulls were studied. There were no significant differences in these factors between steers castrated at approximately one month of age and those castrated after weaning at approximately seven months of age. Bulls gained significantly faster than steers and required less feed per unit of gain. The bulls were not as well finished as the steers at time of slaughter which influenced the carcass characters studied. The steers had a significantly higher dressing percentage and lighter hides. Bull carcasses graded significantly lower, had less fat trim and a higher proportion of edible meat. When based on an appraised live market value the bull carcasses yielded edible meat of comparable quality at a cost of ten to twelve cents per pound less than steers.

Literature Cited

Annual Report. 1946. The effect of castration and hormone administration upon carcass beef production. Fifty-sixth Annual Report Ariz. Agr. Exp. Sta. Pp. 28–30.

Annual Report, 1947. The effect of castration and hormone administration upon carcass beef production. Fifty-seventh Annual Report Ariz. Agr. Exp. Sta. Pp. 33–35.

Bratzler, L. J., R. P. Soule, Jr., E. P. Reineke and Pauline Paul, 1954. The effect of testosterone and castration on the growth and carcass characteristics of swine. JOURNAL OF ANIMAL SCIENCE 13:171.

Deatherage, F. E. and Walter Reiman. 1946. Measurement of beef tenderness and tenderization of beef by the Tenderray Process. Food Research 11:525.

Hunt, Wells E., DeVoe Meade and B. E. Carmichael, 1938. Effect of castration of lambs on their development and quality of meat. Maryland Agr. Exp. Sta. Bul. 417.

THE EFFECT OF AGE AND GRADE ON THE COLLAGEN AND ELASTIN CONTENT OF BEEF AND VEAL [1]

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THE palatability of meat depends upon its tenderness, juiciness, and flavor. Of these three palatability factors, tenderness seems to be the most important to consumers. The first published work showing a relationship between the amount of connective tissue and the tenderness of meat was done by Lehmann in 1897. Mitchell *et al.* (1927) also found that differences in tenderness between beef cuts of the same animal were largely due to collagen and elastin which are the principal proteins in connective tissue. They found that the muscles containing the least collagen were the most tender cuts. The percent of elastin was much more variable among muscles but was found in lesser amounts in the more tender muscles. Other investigators have found that differences in connective tissue content were related to the age of the animal. Mackintosh *et al.* (1936) found meat from yearling steers to contain less collagen and to be more tender than that from mature steers. Mitchell and Hamilton (1933) also reported a slight increase in collagen and a decrease in tenderness as steers grew from baby beef to yearlings. Paul and McLean (1946) however observed large amounts of connective tissue within and between muscles of veal calves. In still other experiments by Mitchell *et al.* (1928) and Bate-Smith (1948) collagen and elastin were not correlated with age and grade of the animal.

The methods of analysis for collagen and elastin which have been used in meat research are for the most part subject to considerable variation. The analyses have in addition varied with respect to the age and grade of the animal and the muscle used. These discrepancies in methods and materials mean that the connective tissue data for ages, grades, and cuts are of little comparative value. It was for these reasons that it seemed desirable to make a study to determine how the connective tissue content in a specific muscle was influenced by age and grade.

[1] Published with the approval of the Director of the Wisconsin Agricultural Experiment Station, Madison.

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Experimental Procedure

Samples of meat from the rib eye (longissimus dorsi) between the 12th and 13th rib were taken from 88 carcasses for this study. Eight samples of each grade were taken from cow carcasses of the U. S. Commercial, Utility, and Cutter grades; steer carcasses of the U. S. Prime, Choice, Good, and Commercial grades, and veal carcasses of the U. S. Choice, Good, Commercial, and Utility grades. Only those cow carcasses which showed completely ossified buttons (cartilage tips on spinous processes) were used.

The lean area of the rib eye was trimmed of external fat and connective tissue, ground three times, wrapped in locker paper, and frozen until time of analysis.

The method for determining collagen and elastin was a modification of the method of Lowry *et al.* (1941), in which a 1.5 gram sample was used and 0.1N alkali was employed to extract the soluble protein. After washing to a negative Biuret test, the collagen was solubilized by autoclaving for 6 hours at 15 pounds pressure. The mixture was filtered into a 50 ml. volumetric and washed with boiling water. One milliliter aliquots of the filtrate were used for nesslerization as outlined by Umbreit *et al.* (1945). The residue from the filtration was determined as elastin using a Kjeldahl digestion followed by nesslerization of an appropriate aliquot of the digest.

Moisture and fat were determined and the collagen and elastin expressed on a fresh and dry fat-free basis.

The data were analyzed by the analysis of variance using the arc-sin transformation recommended by Snedecor (1950).

Results and Discussion

A summary of the data is presented in table 1. Considerable variation existed in the percentage of collagen between animals of the same grade and age. There appears to be no consistent increase or decrease in collagen associated with the grade or the amount of intramuscular fat.

The analysis between ages showed a significantly greater amount of collagen in the veal samples than in the cow or steer samples, with no significant difference between the latter two groups. The average percentages of collagen on a dry fat-free basis for the cows, steers, and veal were 1.84, 1.89, and 2.96 percent respectively, and showed about 50 percent more collagen in the veal than in the other carcasses.

This finding is contrary to the common belief that the amount of collagen increases in older animals and accounts for much of the increase in toughness. The average percentages of collagen are in relative agreement with those obtained by Bate-Smith (1948). She expressed the collagen as a percentage of the coagulable nitrogen and found an average of 5.7, 6.8, and 13.5 percent for cows, steers, and

TABLE 1. SUMMARY OF COLLAGEN, ELASTIN, MOISTURE, AND FAT ANALYSIS OF LONGISSIMUS DORSI OF BEEF ANIMALS OF DIFFERENT AGES AND GRADES

		Collagen %		Elastin %			
Age	Grade	Fresh Wt. Basis	Dry Fat-Free Basis	Fresh Wt. Basis	Dry Fat-Free Basis	Moisture %	Fat %
Aged Cows	Commercial	.424	1.90	.115	0.52	73.19	4.59
	Utility	.392	1.75	.103	0.46	74.14	3.51
	Cutter	.417	1.88	.108	0.49	75.81**	2.06**
	Av.	.411	1.84	.109	0.49	74.38	3.39
Steers	Prime	.418	1.99	***	***	66.65	12.48
	Choice	.375	1.69	.138	0.62	70.97**	6.72*
	Good	.428	1.86	.113	0.49	73.37	3.35**
	Commercial	.445	2.00	.103	0.46	74.38	3.43
	Av.	.416	1.89	.118	0.52	71.34**	6.49**
Veal	Choice	.639	2.78	.269	1.17	76.29	0.79
	Good	.736	3.20	.171	0.74	76.50	0.57
	Commercial	.663	3.02	.256	1.17	77.51	0.40
	Utility	.603	2.83	***	***	78.07	0.51
	Av.	.661**	2.96**	.232**	1.03**	77.09**	0.57**

* Significant at 5% level.
** Significant at 1% level.
*** Analysis not completed.

veal respectively. She also found considerable variation within each grade. Mitchell *et al.* (1927) also found a large variation in the collagen content of the rib eye of carcasses of equal grade, reporting an average of 6.1 percent collagen on a nitrogen basis with a range of 2.5 to 9.4 for four animals. A year later Mitchell *et al.* (1928) reported the percent collagen in the rib eye of 12 carcasses ranging from choice steers to a canner cow. Again the collagen content was not related to grade or age in this group of mature animals. Paul and McLean (1946) observed, in a histological study of veal, that the muscles of the round contained much collagenic connective tissue and the elastic connective tissue was very abundant. While a quantitative study of these constituents was not made, this observation is in accord with those of Bate-Smith as well as the present study.

Since a number of workers have shown that differences in tenderness between muscles of the same carcass are related to their collagen content, it would appear that the collagenic connective tissue of veal has different physical or chemical properties. Bray *et al.* (1951) showed that the connective tissue of veal had a higher moisture content than that of older animals. It might be postulated that the more hydrated connective tissue would be more easily hydrolyzed during the cooking process and therefore have less effect upon the tenderness of the meat. Their results showed, however, that the veal connective tissue was hydrolyzed at about the same rate as that from steers but there was less hydrolysis in the connective tissue of old cows.

The analysis for elastin also showed coniderable variation between animals of the same grade and age and further that the difference between grades was not significant. The percentage of elastin in the cow samples (0.49) was not significantly different from that of the steers (0.52), but both the steer and cow samples contained significantly less elastin than the veal (1.03 percent). The chemical determinations of elastin in the muscles of beef animals by Mitchell *et al.* (1927) (1928) (1933) and Harrison *et al.* (1949) were the only values found in the literature. In neither case was there a correlation between the elastin content and the grade of the animal. Mitchell *et al.* (1928) did find, however, that samples from two steers of the medium grade contained an average of 0.21 percent elastin nitrogen, while similar samples from comparable steers slaughtered 213 days later contained only 0.02 percent. Similarly, Harrison *et al.* (1949) found a greater amount of elastin in the beef from a yearling steer than that from an 8-year-old cow.

Two possible explanations for the higher amount of collagen and elastin in veal seem justified.

It is known that collagen fibers are surrounded by reticulum, which is either present in greater amounts or is more resistant to mild alkali in the young animal (Lloyd *et al.*, 1935, 1936, 1933). Furthermore, many workers believe that the reticulum is determined as collagen in most cases (Gersh, 1951). If a greater amount of reticulum is associated with the collagenic fibers in the young animals and the reticulum is not a factor in determining tenderness, the higher values for veal might be expected.

A number of investigators have shown that the muscle fiber size increases with maturity (Hammond, 1932; Brady, 1937; Hiner, 1953). McMeekan (1940–41) has shown in hogs that from birth until 28

weeks of age, the muscle fibers increase over eight times in diameter and 70 times in cross-section area, thus increasing the amount of intracellular protein. At the same time, it is the opinion of some workers that the absolute amount of connective tissue remains relatively constant throughout life, and that small connective tissue fibers coalesce into stronger fibers with age (Gersh *et al.*, 1949; Porter, 1951). Assuming the above to be true, any given size of sample from an older animal, possessing larger fibers, will contain more intracellular protein than a sample of veal of similar size. A similar explanation could account for the higher percent of elastin in veal.

Summary

Data have been secured to determine the effect of age and grade on the collagen and elastin content of the longissimus dorsi of beef animals. The samples for analysis were selected from three grades of cow carcasses, four grades of steer carcasses, and four grades of veal carcasses.

Considerable variation in the collagen and elastin content was found within grades for each age group and in no case were the differences in these measurements between grades significant. There was no difference between the collagen and elastin content of samples from steers and cows, but the samples from both of these older groups contained less collagen and elastin than veal.

Because veal is more tender than meat from more mature animals, it is concluded on the basis of these data that the percentage of collagen and elastin in the longissimus dorsi of beef animals is not a critical measure of tenderness of meat.

Literature Cited

Bate-Smith, E. C. 1948. Observations on the pH and related properties of meat. Jour. Soc. Chem. Ind. 67:83.

Brady, D. E. 1937. A study of factors influencing tenderness and texture of beef. Proc. Amer. Soc. Animal Prod. pp. 246–250.

Bray, R. W., P. H. Phillips and G. Bohstedt. 1951. Moisture, ash, and fat content of connective tissue from cattle of different ages. JOURNAL OF ANIMAL SCIENCE 10:394.

Gersh, I. and H. R. Catchpole. 1949. The organization of ground substance and basement membrane. Amer. Jour. Anat. 85:457.

Gersh, I. 1951. Some functional considerations of ground substance of connective tissues. 2nd Conference on Connective Tissues, 11–14. Josiah Macy Jr. Found.

Hammond, J. 1932. Growth and development of mutton qualities in the sheep. Oliver and Boyd Pub. Co., London.

Harrison, D. L., B. Lowe, B. A. McClurg and P. S. Shearer. 1949. Physical, organoleptic and histological changes in three grades of beef during aging. Food Tech. 3:284.

Hiner, R. L., O. G. Hankins, H. S. Sloane, C. R. Fellers and E. E. Anderson. 1953. Fiber diameter in relation to tenderness of beef muscles. Food Res. 18:364.

Lehmann, K. B. 1897. Cited by Mitchell *et al.* 1927.

Lloyd, D. J., R. H. Marriot and W. B. Pleass. 1933. The swelling of protein fibers. Part I. The swelling of collagen. Trans. Farad. Soc. 29:554.

Lloyd, D. J. and R. H. Marriot. 1935. The swelling of structural proteins. The influence of the reticular tissue on the swelling of collagen in water and hydrochloric acid. Proc. Roy. Soc. B. 118:439.

Lloyd, D. J. and R. H. Marriot. 1936. The swelling of protein fibers. Part V. The swelling of single collagen fiber bundles under load. Trans. Farad. Soc. 32:932.

Lowry, O. H., D. R. Gilligan and E. M. Katersky. 1941. Determination of collagen and elastin in tissue. Jour. Biol. Chem. 139:795.

Mackintosh, D. L., J. L. Hall and G. E. Vail. 1936. Some observations pertaining to tenderness of meat. Proc. Amer. Soc. Animal Prod. pp. 285–289.

McMeekan, C. P. 1941. Growth and development in the pig with special reference to carcass quality characteristics. Cambridge University Press, London.

Mitchell, H. H., R. L. Zimmerman and T. S. Hamilton. 1927. The determination of the amount of connective tissue in meat. Jour. Biol. Chem. 71:379.

Mitchell, H. H., T. S. Hamilton and W. T. Haines. 1928. Some factors affecting the connective tissue content of beef muscle. Jour. Nutr. 1:165.

Mitchell, H. H. and T. S. Hamilton. 1933. Effect of long continued exercise upon the chemical composition of the muscles and other tissues of beef cattle. Jour. Agr. Res. 46:917.

Paul, P. and B. B. McLean. 1946. Studies on veal. I. Effect of different internal temperatures on veal roasts from calves of three different weights. Food Res. 11:107.

Porter, K. R. 1951. Repair processes in connective tissues. 2nd Conference on Connective Tissue. 126–158. Josiah Macy Jr. Found.

Snedecor, G. W. 1950. Statistical methods, 4th ed. Iowa State Col. Press, Ames, Iowa.

Umbreit, W. W., R. H. Burris and J. F. Stauffer. 1945. Manometric techniques and tissue metabolism. Burgess Pub. Co.

SOME SOURCES OF ERROR IN WEIGHING STEERS OFF GRASS

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EXPERIMENTAL studies with cattle on pastures or ranges usually use weight changes as a measure of the response to a condition or treatment. The weight taken at any time includes the actual tissue weight of the animal plus the contents of the digestive tract (fill). Under normal conditions the tissue weight of an animal should not show large day to day variations. The amount of fill, however, may be quite variable, increasing as the animal eats or drinks, and decreasing with the excretion of body wastes. Lush *et al.* (1928) observed that experimental error may be rather closely related to the size and capacity of the animal's digestive tract. The differences noted in weighing animals include differences due to tissue weight, fill, and weighing errors. The latter two of these sources of variation must be minimized if experiments are to yield highly repeatable results.

Errors in weighing can be minimized by using more accurate scales and by more care on the part of the scale operator in preventing mistakes. Errors due to differences in fill are more difficult to control. If errors due to fill are random, with equal likelihood of affecting any animal of any group to the same extent, then the net result of the variations in fill will be seen throughout the data and upon analysis will increase the unmeasured variation components. Such effects tend to increase standard errors of estimates and error mean squares, and reduce the magnitude of estimated correlation coefficients. If, on the other hand, fill differences are not random, they may cause any number of effects. Of concern in this paper is the effect that fill differences may have on lot means.

Materials and Methods

The data for this study include the weights of 180 steers which were being used in a series of management studies. Unless otherwise indicated, the studies reported herein were made during the interim

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between management studies, at which time the steers were not allotted to other experiments but were under a common system of management.

Randomly selected groups of the steers were subjected to different conditions and their response studied. To do this, a base weight was established for each steer, either by averaging three shrunk weights or by taking one weight under conditions of more extreme shrinkage. In obtaining data the steers were weighed to the nearest five pounds on recording type scales. The weight change from the base weight to the weight under the condition being studied was the unit used for analysis.

Results and Discussion

Source of Fill

The first phase of the study was concerned with attempting to determine whether feed or water was the greatest source of variation in fill. The base weight was an average of three shrunk weights taken May 19, 21 and 23, 1952. The steers weighed an average of about 820 lbs. (st. dev. of 60). A component of variance analysis indicated that about 95 percent of the variation in weights was due to differences in the mean weights among the steers. Patterson's (1947) data indicated a greater percentage of the overall variation was due to differences among the steers. The other five percent of the variation was due to the failure of the steers to weigh the same at each weighing. This latter variation is interpreted as being largely due to fill differences, although it also contains variations due to rounding off.

The steers were divided at random into three groups during the late afternoon of June 10. Group 1 was placed in a dry lot, Group 2 remained on native grass pasture but was not allowed access to water, while Group 3 was left on pasture with water. The following morning, these groups were gathered and the steers weighed as rapidly as possible consistent with good weighing technique. As the steers were weighed they were further separated, one half of each group going to a dry lot and the other half going to pasture and water. Four to six hours later the steers were reweighed. It was observed that the steers that were turned out to pasture drank considerable water, but did no appreciable amount of grazing.

This procedure was repeated on July 6 and 7 when the vegetation, though plentiful, was much lower in moisture. Chance alone determined the treatment imposed on either of these weigh days. Table 1

shows the weight changes that occurred between the base weights and the first weights of each weigh day.

The weight changes for the three groups of steers taken on June 11 were significantly different. Apparently the steers that were on pasture, but without water, ate enough to make their fill greater (about 15 lb.) than the steers kept in the dry lot. The steers that were left on pasture with water must have taken on both water and feed. They had about

TABLE 1. WEIGHT CHANGES SHOWN BY STEERS FROM A BASE SHRUNK WEIGHT TO WEIGHTS TAKEN UNDER OTHER CONDITIONS

Group	Condition	5/21 to 6/11 No. Steers	5/21 to 6/11 Wt. Change	5/21 to 7/7 No. Steers	5/21 to 7/7 Wt. Change
			(lbs.)		(lbs.)
1	Overnight in dry lot	50	38.5	45	103.6
			(326)		(318)
2	Overnight on pasture (no water)	49	54.1	49	101.0
			(376)		(426)
3	Overnight on pasture	49	94.7	49	125.5
			(467)		(383)

( )Indicates variance of differences.

56 lb. more fill than the shrunk steers and 40 lb. more fill than the steers on pasture without water. These data indicate that water was by far the greater source of fill, but that green feed was also important.

The July 7 weight changes showed different results. The steers that had access to pasture but were without water showed no more fill than the dry lot steers. The group that was on pasture with water, however, had about 23 lb. more fill than the average of the other two groups. In this case, water seemed to be the only source of fill differences between groups.

The Effect of Subsequent Treatment on Different Amounts of Fill in Steers

As stated previously, the steers were divided into two groups after the first weighings on June 11 and July 7. The method of accomplishing this consisted of turning alternate groups of five into a dry lot with the other groups going back to pasture. All steers going to pasture were mixed regardless of previous treatment, as were also the steers that went into the dry lot. Tables 2 and 3 show the results of this portion of the study.

Weight changes 1 and 2 are the differences between the base weights and the first and second weights respectively taken on the indicated days. In the weight change 1 column, lines A1 and B1 show different estimates of the same population mean as do lines A2 and B2 and lines A3 and B3. The double estimates were combined to give the estimates shown in table 1. The treatment effect is the difference

TABLE 2. EFFECT OF SUBSEQUENT TREATMENTS ON STEERS WITH DIFFERENT AMOUNTS OF FILL

(June 11)

Treatment	No. Steers	Wt. Change 1	Wt. Change 2	Treat. Effect	Time
		lb.	lb.	lb.	Minutes
A. Shrunk after:					
1. O/n dry lot	25	42.2(390)	19.6(350)	−22.6**(52)	240
2. O/n pasture (no water)	24	57.7(315)	35.6(150)	−22.1**(87)	210
3. O/n pasture	23	95.3(462)	59.6(299)	−35.7**(92)	285
B. To pasture and water after:					
1. O/n dry lot	25	34.8(247)	89.4(439)	54.6**(187)	290
2. O/n pasture (no water)	25	50.6(425)	100.2(508)	49.6**(514)	260
3. O/n pasture	26	94.2(486)	99.0(529)	4.8 (559)	335

O/n Indicates over-night.
( ) Indicates variance of differences.
** Indicates probability of chance occurrence less than .01.
Wt. Change 1 shows the weight change from the base weight to the first weight on the indicated day.
Wt. Change 2 shows the weight change from the base weight to the second weight on the indicated day.

between the preceding columns and summarizes the effect of either further shrinking or allowing the animals to take on fill for the average time shown in the last column. The variance estimates shown were calculated from the items that gave the mean estimates.

The group of steers that had been shrunk overnight (Line A1, table 2) continued to shrink during the next four hours at the rate of slightly less than one lb. every ten minutes. A study of the weight changes in line A2 indicates that the steers that had overnight access to pasture but no water shrunk the following day at a rate slightly in excess of a pound every ten minutes. The full group (line A3) had a slightly greater shrinking rate than either of the other groups during the shrinking period on June 11. The comparison is not statistically significant, even though *a priori* one might suspect that it is real. A study of lines B1, B2, and B3 indicates that the steers that went to pasture and water tended to regain weight in proportion to the loss shown in the previous treatment.

Table 3 shows that in July the rates of shrinkage were very much the same as in June except that the overnight on pasture with water group shrunk relatively slower than before.

It should be remembered that at the time of the July 7 weights (table 1) there appeared to be no difference between the fill contained by steers shrunk in dry lot and those left on grass overnight but

TABLE 3. EFFECT OF SUBSEQUENT TREATMENTS ON STEERS WITH DIFFERENT AMOUNTS OF FILL

(July 7)

Treatment	No. Steers	Wt. Change 1	Wt. Change 2	Treat. Effect	Time
		lb.	lb.	lb.	Minutes
A. Shrunk after:					
1. O/n dry lot	21	106.2(235)	73.6(218)	—32.6**(29)	360
2. O/n pasture (no water)	25	98.8(405)	70.2(350)	—28.6**(30)	225
3. O/n pasture	25	123.0(309)	89.8(318)	—33.2**(41)	315
B. To pasture and water after:					
1. O/n dry lot	24	101.4(393)	152.2(507)	50.8**(230)	445
2. O/n pasture (no water)	24	103.2(456)	154.0(341)	50.8**(299)	345
3. O/n pasture	24	128.1(462)	139.4(375)	11.3 (270)	405

O/n Indicates over-night.
( ) Indicates variance of difference.
** Indicates probability of chance occurrence less than .01.
Wt. Change 1 shows the weight change from the base weight to the first weight on the indicated day.
Wt. Change 2 shows the weight change from the base weight to the second weight on the indicated day.

without water available. There was also less difference between these two groups and the group that had been on pasture with water than was found among the comparable groups at the time of the June 11 weights.

If the shrinking of steers tends to equalize the fill, the variances within groups should be reduced. The variances of the first and second weight changes taken on June 11 (from line A1, table 2) were 390 and 350 respectively. These variances are the variances of differences and equal the variance of the first weight "σ_x^2" plus the variance of the second weight "σ_y^2" minus twice the covariance of the two weights "$\rho_{xy}\sigma_x\sigma_y$" (Snedecor, 1948). The first of these variances is fixed when the sample of steers is chosen. The second variance is related to the first in that the same animals are involved but may differ because of changes in the tissue weights of the animals and also because of different amounts of fill. The correlation between the two weights should be largely due to the tissue weight of the animal.

The chosen variances (390 and 350) have two common components, i.e., σ_x^2 and ρ_{xy}. They differ in their contribution to the variance of the June 11 weight σ_y^2. The shrinking on June 11 reduced the variance as was hypothesized. Since the value 390 was the variance of the difference of two shrunk weights it would not be expected to be reduced very much by further shrinking.

By making similar comparisons of the variances for the other June 11 weight changes in lines A2 and A3 and it will be noted that the variances were appreciably reduced. The comparable variance changes for the July 7 weights (table 3) were not large and one actually increased. The latter does not refute the hypothesis because such a slight change could be explained as a result of rounding off if in actuality there was no change in variance.

Another approach can be made to this problem. If the animals that gain more between the two weights do this partially by taking on more fill and if these animals that have the most fill lose more during a subsequent shrinking period, there should be a negative correlation between the gain from the base weight to the first weight on either of the weigh days and the loss during a subsequent shrinking period. Table 4 shows the estimated correlation coefficients between these weight changes.

These data seem to support the hypothesis that shrinking tends to equalize the fill in steers weighed off pasture. The greatest advantage appears to be with steers weighed off green, actively growing pastures.

Lot Variations in Fill

During the winter period of 1952–53 the steers were equally divided among ten pastures for a series of studies on mineral and protein supplementation. On the afternoon of March 9, 1953, the steers were gathered and a full weight was taken immediately. The steers were shrunk over night in dry lots and reweighed the following morning. An analysis of variance on the differences between these full and shrunk weights indicated that average lot shrinkages varied more than could be explained by the average variation within lots. The calculated F value was highly significant. Neither treatments to which the animals had been subjected nor any other known sources of variation would satisfactorily explain the shrinkage differences among lots. It was concluded that the differential shrinkages were probably due to the various lots having different amounts of fill at the time that they were gathered.

TABLE 4. ESTIMATED CORRELATION COEFFICIENTS BETWEEN GAIN FROM BASE WEIGHTS AND LOSS DURING SHRINKING

	Group		
	A1	A2	A3
June 11	—.324	—.764**	—.626**
July 7	—.275	—.387	—.144

** Indicates probability of chance occurrence less than .01.

Whether the differences between lots were due to different grazing habits or to different groups having watered more recently is not known. The steers were all run in a common pasture under uniform conditions until May 1st and 2nd at which time individual full and shrunk weights were again taken. A similar analysis was run on weight differences, using the same groups that had existed during the winter period. As would be expected in a uniformity trial, the mean square for lot shrinkages was comparable to the mean square for within lot shrinkages.

The March 10th weight was used as a base weight in studying the rate of shrink in a series of studies during the following two months. On March 28th, the steers were gathered and driven to the corral about 8:00 a. m. and divided at random into four dry lots. The lots were weighed at about one and one-half hour intervals to get a measure of the rate of weight loss by steers being held undisturbed in a dry lot. Table 5 gives the results of this trial.

On April 2nd the steers were again gathered at about the same time of day and this time weighed at about one hour intervals, with the unweighed group being driven up and down a lane while waiting to be weighed. In pasture studies it is frequently necessary to drive some lots of animals several miles to the scales while other lots may be pastured near the weighing facilities. The results of this trial are shown in table 6.

TABLE 5: WEIGHT CHANGES OF GROUPS OF STEERS FROM BASE WEIGHTS TO WEIGHTS TAKEN AFTER VARYING LENGTHS OF TIME IN DRY LOT

Time Weighed	No. Steers	Wt. Change (lb.)	Variance Estimate
8:20– 9:20	42	17	445
9:50–10:25	43	6	789
11:20–12:05	43	—6	576
1:10– 2:00	47	—7	395

Tables 5 and 6 show a consistent trend for steers to lose weight while being held in dry lot or being driven. The purpose of these trials was to determine estimates of the rate of weight loss under these conditions and to compare these rates. The groups reported in table 5 were weighed over a longer period of time than those in table 6. If rate of shrinkage (weight loss) is curvilinear in time as it must be, then, other

TABLE 6. WEIGHT CHANGES OF GROUPS OF STEERS FROM BASE WEIGHTS TO WEIGHTS TAKEN AFTER HAVING BEEN DRIVEN VARYING LENGTHS OF TIME

Time Weighed	No. Steers	Wt. Change (lb.)	Variance Estimate
8:05– 8:50	45	18	633
9:05– 9:35	46	15	486
10:00–10:30	47	4	431
11:00–11:35	38	1	644

factors being equal, the groups of steers reported in table 5 might be expected to show a slower rate of shrinkage than those in table 6. This proved to be the case in these trials. However, if one considers only the first three groups of steers that were held in dry lot and compares their rate of shrinkage to the rate shown by the groups that were driven (table 6), the time periods are comparable. If a linear rate of shrinkage is assumed during these three hour periods, the rate of shrinkage was .148 lb. per minute for the dry lot steers and .109 for the steers being driven. It is believed that the shrinkage rate for the dry lot steers was over-estimated. When the whole weight period was considered for the dry lot steers the linear rate of shrinkage was .086 lb. per minute. A rate that is somewhere between these (.086 and .148) is believed to be a better estimate. More significant, perhaps, is that these results give one no reason to believe that driven steers will shrink faster than those being held in a dry lot.

The next phase of the study was concerned with trying to determine if time of gathering was related to the amount of fill in the animals. Under the system of management most commonly used at this station, cattle are gathered for weighing during the morning and weighed as soon as possible. On April 14th one-third of the steers were gathered before they had commenced grazing and were weighed immediately. One-half of the remaining steers were gathered and weighed about two hours later. The remaining steers were brought in and weighed after another two hours. The results of this study are shown in table 7.

TABLE 7. THE EFFECT OF TIME OF GATHERING ON AMOUNT OF FILL

Time Weighed	No. Steers	Wt. Change (lb.)	Variance Estimate
5:25– 6:15	58	68	901
7:35– 8:40	60	50	928
9:45–10:40	61	62	1054

By 7:30 a. m. a dust storm was in progress. Since weather can affect both rate of shrinkage and grazing habits, it was decided to repeat this trial on April 25th. These results are presented in table 8.

These results suggest that the amount of fill in the animals varied with the time gathered. The steers were driven about two miles before being weighed. It would seem likely that the loss during the first two hours was the result of the animal excreting at a faster rate than he was eating. During the next two hours he apparently grazed at a much faster rate than he excreted his body wastes, so that he approached or exceeded his first weight. These responses approach significance at the .05 level but are not conclusive. During the Fall of 1953, the experiment was repeated using 110 beef cows that weighed about a thousand pounds each. They were gathered at two different times and the second group followed the first by about two and one-half hours. Here, again, the group that was gathered before they could graze apparently had more fill than those that were gathered later. In this instance the difference was significant at the .05 level.

Application of Results

The purposes of this study were to determine, if possible, what causes fill differences, how fill differences are distributed, and what precautions should be taken to reduce their detrimental effects on weight data. The steers used were available during certain seasons and could be managed in such a way that responses under known conditions could be studied. The results of this study with steers are

TABLE 8. THE EFFECT OF TIME OF GATHERING ON AMOUNT OF FILL

Time Weighed	No. Steers	Wt. Change (lb.)	Variance Estimate
5:30–6:00	48	115	1148
7:30–8:15	60	105	811
9:25–9:55	68	129	812

believed to indicate some of the causes of error that affect the weights of all classes of cattle, even though the extent of the effects may not be indicated. Also, the pasture conditions that existed in this study may cause the extent of the measured responses to be different than those under other conditions.

Many experiments are designed to measure, or determine the existence of very small weight responses. If these experiments are to yield unbiased results, every precaution must be taken to assure exacting weight records. This study has indicated that differences in fill can either increase the overall variation so that statistical significance of results is more difficult to obtain, or that group means may be biased in such a manner that the repeatability of results may be appreciably reduced.

Summary

One hundred and eighty head of yearling and two-year-old steers were used over a period of 15 months to study the variations that occur in the amount of fill contained by steers running on pasture. The weight changes between a base weight taken under uniform conditions and subsequent weights taken under controlled conditions constituted the items of study.

Under the conditions of the experiment it was found that the availability of water was a more important source of fill variation than the availability of pasture grass. There was an indication that under some conditions the differences in amount of fill among the animals tend to be lessened by shrinking. Animals in different pastures exhibited different amounts of fill when gathered during the afternoon. Using full weights under such conditions would tend to reduce the repeatability of experiments. Steers weighing from eight hundred to a thousand lb. lost about one lb. every ten minutes during the first three to four hours of shrinking whether they were held in a dry lot or were being quietly driven. Animals on pasture early in the morning appeared to lose fill faster than it was replaced.

These findings emphasize the necessity for using standardized procedures in weighing cattle. It is suggested that all lots of cattle should have water equally available at all times. Shrinking cattle for a period of 10 to 15 hours before taking the weights that are to be used in the final evaluation of treatments effects may increase experimental efficiency. If several lots are involved, they should be weighed as rapidly as possible to prevent excessive loss of fill by those being weighed last.

Literature Cited

Lush, J. L., F. W. Christensen and W. H. Black. 1928. The accuracy of cattle weights. Jour. Ag. Res. 36:551.

Patterson, R. E. 1947. The comparative efficiency of single versus three-day weights of steers. JOURNAL OF ANIMAL SCIENCE 6:237.

Snedecor, Geo. W. 1948. *Statistical Methods*, 4th Edition. The Iowa State College Press. Ames, Iowa.

WEANING WEIGHTS OF HEREFORD CALVES AS AFFECTED BY INBREEDING, SEX, AND AGE

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IT was the purpose of this study to derive a series of estimates, indicative of the quantitative effect of various environmental and genetic factors on the weaning weights of 546 Hereford calves. These estimates were used to adjust weaning weights so that weaner calves and cows could be compared as to their relative merit for herd replacements. Specifically, the environmental factors studied were year of birth, age of dam, and age of calf at weaning. The genetic factors were inbreeding of dam, inbreeding of calf, and sex of calf.

Materials and Analysis

The data employed concerned the purebred Herefords at the San Juan Basin Experiment Station, Fort Lewis A & M College, Hesperus, Colorado. They were confined to the analysis of the weaning weights of 546 conventional type Hereford calves, out of conventional type Hereford dams. Comprest cattle used in type experiments were excluded. The calves were dropped during the years 1946–1951, inclusive.

In 1946, an inbreeding program was initiated in which several lines were established. Only line bulls were used as sires. Frequent sire-daughter matings were made in an attempt to raise the level of inbreeding within lines rapidly. Selection was practiced among dams but not among calves. Inbred bulls from the various lines were progeny tested on outbred herds.

The range land at Fort Lewis is typical of the San Juan Basin of southwestern Colorado. The temperature in degrees Fahrenheit varies from a maximum of 99 to a minimum of −32 with an average annual precipitation of 18.29 inches (Yearbook of Agriculture, 1941). The altitude is approximately 8,870 feet above sea level (Gannett, 1906). Calving started in March and generally ended by late June. Soon after birth, all male calves from type test matings were castrated and de-

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horned. All the female calves were dehorned. All calves remained with their dams on range until they were weaned in late November or early December. At this time, each calf was weighed once. The type test was concluded with the 1950 calves, hence there were no steer calves in 1951.

For each year under investigation, dams were divided into groups according to their ages at calving. The weaning age of the calf was given in days. Only the unadjusted weaning weights were used in obtaining the estimates of the constants under study. Inbreeding coefficients were computed by use of Wright's formula as presented by Lush (1945). In 1946, the average inbreeding in the herd was 5%. This value was assigned to all calves and dams in 1946 and thereafter to all calves resulting from type test matings.

The data, the distribution of which is shown in table 1, were arranged in seventeen simultaneous equations using the principle of least squares as described by Hazel (1946). The absorption-iterative method (Henderson, 1948) was used to estimate the various parameters. Significance of the various categories has been shown in the variance analysis based on the method used by Hazel (1946). The details and mechanics of setting up the equations, solving them, and developing the variance study have been described in their entirety by Landblom (1954).

The solution of the system of equations as described in detail by Landblom (1954) resulted in the estimates for the coefficients presented in table 2. Table 3 shows the variance study made for the weights of the calves. It may be noted that significance at the .05 or .01 levels was found for all categories.

The constants derived from the data of the previous six year period were tested on the weaning weights of the 1952 and 1953 calves in the Fort Lewis herd. The hypothesis would be that their application should remove, except for sampling error, those differences between calves due to age of calf and dam, inbreeding of calf and dam, and sex. Thus, differences between weaning weights of inbreds and outbreds, heifers and bulls, of varying ages and from varying ages of dams should not be significant if the constants are applicable.

The data on which comparisons of inbreds vs. outbreds and males vs. females have been made are shown in table 4. The mean weights of these two comparisons are shown before and after adjustment or correction. The means of adjusted weights of bulls vs. heifers differed by a small amount which was not significant. Thus, it appeared that

TABLE 1. NUMBERS OF CALVES IN VARIOUS CATEGORIES AND AVERAGE WEANING AGES

Sex		Steers						Bulls							Heifers							
Year		46	47	48	49	50	n	46	47	48	49	50	51	n	46	47	48	49	50	51	n	N
	2	—	1	3	3	6	13	1	3	1	4	7	7	23	4	7	5	8	13	7	44	80
Age	3–5	8	19	6	10	8	51	5	2	14	27	28	33	109	15	8	18	22	21	25	109	269
of	6–8	7	7	7	7	6	34	8	10	4	8	5	12	47	11	10	6	1	5	12	45	126
Dam	9+	4	1	1	5	4	15	1	6	3	7	3	2	22	4	9	8	4	7	2	34	71
	n	19	28	17	25	24	113	15	21	22	46	43	54	201	34	34	37	35	46	46	232	546
	0–10	19	28	17	25	24	113	15	12	10	18	10	31	96	34	19	17	14	17	23	124	333
	11–20	—	—	—	—	—		—	8	4	13	7	11	43	—	13	12	9	9	7	50	93
Fx*	21–30	—	—	—	—	—		—	1	5	9	14	8	37	—	2	5	7	10	10	34	71
of	31–40	—	—	—	—	—		—	—	3	6	10	3	22	—	—	3	5	7	4	19	41
Calf	41–50	—	—	—	—	—		—	—	—	—	2	1	3	—	—	—	—	3	2	5	8
	Ave. %	5	5	5	5	5	—	5	10	16	17	22	13	—	5	10	14	16	19	15	—	—
	0–10	19	28	17	25	24	113	15	20	17	38	25	33	148	34	32	32	27	30	28	183	444
Fx	11–20	—	—	—	—	—		—	1	3	6	15	16	41	—	2	3	7	12	11	35	76
of	21–30	—	—	—	—	—		—	—	2	2	3	3	10	—	—	2	1	2	3	8	18
Dam	31–40	—	—	—	—	—		—	—	—	—	—	2	2	—	—	—	—	2	4	6	8
	Ave. %	5	5	5	5	5	—	5	6	8	7	10	10	—	5	6	7	8	10	11	—	—
Ave. weaning age		216	211	201	209	211	210	218	225	206	215	204	212	213	214	219	200	216	211	207	211	211

*Fx = Inbreeding coefficient in per cent

the sex correction developed in the study was a good fit for the 1952 and 1953 calf crops. In contrast, the adjustment for inbreeding effect resulted in corrected weights being much greater for the inbreds than for the outbreds. These differences were significant and were in the same direction in each of the two years.

TABLE 2. FINAL ESTIMATES OF CONSTANTS[1]

Variable		Final Estimate of Effect on Weaning Weight in lb.
Years	1946	5.91
	1947	—24.10
	1948	— 2.04
	1949	1.62
	1950	— 1.18
	1951	19.79
Age of dam	2-year-old	—15.15
	3- to 5-year-old	4.68
	6- to 8-year-old	20.71
	9-year-old and over	—10.24
Sex	Steer	—6.24
	Bull	14.34
	Heifer	—8.10
Inbreeding 1% inbreeding of calf		—1.76
1% inbreeding of dam		—1.15
Age of calf at weaning Effect of one day		1.67
W intercept or u		76.36

[1] Average weaning weight for years 1946–1951 inclusive was 402.64 lb.

The inbreeding in the 1952–53 population was almost double that in 1946–51. At higher levels of inbreeding the regression may not be so great. The possibility of curvilinearity of inbreeding effects was then investigated in the 1946–51 data. For calf inbreeding effect, but not for the dams' inbreeding, there was significant deviation from linearity. However, when weight adjustments for curvilinearity were applied to the 1952–53 calf crops even greater differences appeared between the adjusted weights of the inbreds and outbreds than when linear adjustments were used.

Discussion

Previous research has indicated that the weaning weight of a calf is greatly influenced by the age of its dam. Knapp *et al.* (1942) found that maximum weaning weights may be expected from six-year-old cows with a gradual increase from two to six years of age and a more rapid decrease from six to eleven years. Knox and Koger (1945) showed that the age of maximum weight and production of range cows was from six to eight years with the peak at seven years. Sawyer *et al.*

TABLE 3. VARIANCE ANALYSIS FOR WEIGHTS OF CALVES

Variability due to	D/F	Sums of Squares	Mean Square	Observed F
Total	545	2,731,179		
Total reduction	13	1,448,815		
Direct effects				
Year	5	79,116	15,823	6.57**
Age of dam	3	70,040	23,347	9.69**
Sex	2	56,110	28,055	11.64**
Inbreeding of calf	1	89,478	89,478	37.13**
Inbreeding of dam	1	13,424	13,424	5.57*
Age of calf	1	657,016	657,016	272.62**
Residual	532	1,282,364	2,410	

** Significant at .01 level; * Significant at .05 level.

(1948) found that two-year-old heifers weaned calves that were 75 pounds lighter than those from mature cows. Weaning weights increased with increasing age of dam through eight years but then declined. To adjust calf weights to those from 6–8 year old cows Bloom (1953) added 59 pounds to calves from 2 year old cows, 23 pounds to calves from 3–5 year old dams, and 4 pounds to calves from dams of 9 and over.

In general, the papers reviewed showed males to be heaviest in weaning weight. Significant differences in rate of gain prior to weaning have been observed, male calves gaining at a faster rate (Black and Knapp, 1936, Knapp and Black, 1941 and Bloom, 1953). Knapp *et al.* (1942), Koch (1951), and Bloom (1953) found that male calves were significantly heavier at weaning. Koger and Knox (1945) indicated that steer calves significantly outweighed heifer calves at weaning. In contrast, Sawyer *et al.* (1948) found evidence that heifer calves were heavier at weaning than steer calves; however, the difference lacked significance.

TABLE 4. MEANS OF ACTUAL AND ADJUSTED WEANING WEIGHTS OF HEREFORD CALVES IN 1952 AND 1953

Item	Inbred					Outbred						
	1952		1953		Av. or	1952		1953		Av. or	All	All
	Male	Female	Male	Female	Total	Male	Female	Male	Female	Total	Males	Females
Number	18	23	30	27	98	36	29	30	20	115	114	99
Calf age (days)	228	220	205	196	212	224	229	208	196	214	216	210
Dam age (years)	4.4	3.7	4.0	4.0	4.0	4.5	4.3	3.6	4.4	4.2	4.1	4.1
Calf inbreeding %	31	29	31	29	30	5	5	5	5	5	18	17
Dam inbreeding %	21	19	22	19	20	5	5	5	5	5	13	12
Actual weight	395	339	394	370	374	405	395	447	420	417	410	381
Adjusted weight	439	420	483	487	457	395	403	469	468	434	447	444

It has been demonstrated by Koch (1951) that weaning weights were adversely affected by inbreeding of calf and dam.

In a study of influences of calf age on preweaning growth of Hereford cattle, Rollins *et al.* (1952) found a regression of weight on age, for the period birth to four months of 1.90 lb. per day and from four to eight months, 1.81. Koch (1951) found that weight increased 2.27 lb. with each additional day of age.

Primarily, the estimates of the constants in the present study were obtained to furnish a basis for adjusting the weaning weights of calves to be compared within year of birth. In practice, the estimates for year effect would not be used, since each was assumed to be descriptive of the effect of a given year in the past. The use of the estimates would thus be confined to those of age of dam, sex of calf, inbreeding of calf, inbreeding of dam, and age of calf at weaning. Each of these estimates was derived as an average effect over a period of six consecutive years, except steer effect which was based on a five-year record.

The estimates agree fairly well with other studies on age of dam effect. It is likely that some confounding between dams' ages and calves' ages may occur where all weaning takes place at one time and where yearlings are placed in the breeding pasture as they reach 15 months of age. The estimates derived herein should be free from such bias. Sex effect as found in this study agrees in direction but differs in degree from other studies as steers weighed only slightly more than heifers at weannig. Bulls were considerably heavier than steers and heifers at weaning, a phenomenon generally observed.

The very striking effects of inbreeding on weaning weight to a degree substantiate Koch's findings. However, higher levels of inbreeding existed in the present herd than in that described by Koch. There is of necessity some correlation between inbreeding of calf and dam and it is assumed again that the method of analysis derives the estimates independent of this association. Inbreeding effects were very large, for 25% inbreeding of calf and dam would be expected to decrease weaning weight about 73 pounds. It appears from the present study that significant effects on weaning weights may be attributed to inbreeding, sex, and age, and should be considered in comparing calves, dams, and sires. The extent to which the present results may be widely applicable is unknown, but there is considerable agreement between the results found in these data and those reported in other studies in the western United States.

Summary

A set of descriptive constants was developed, whereby the variations in weaning weight caused by inbreeding, age and sex could be estimated in a herd of Hereford beef cattle. These constants were developed to increase effectiveness of selection for weaning weights among calves to be considered for replacements and their dams. The variables considered were years, age of dam, sex of calf, inbreeding of calf and dam, and age of calf at weaning; all were significant in their effect at the 0.05 or 0.01 level. Effect in pounds as deviations from the average weaning weight were:

(1) Years: 1946, +6.; 1947, −24; 1948, −2; 1949, +2; 1950, −1; 1951, +20.

(2) Age of dam: 2 years, −15; 3 to 5 years; +5; 6 to 8, +21; 9 and over, −10.

(3) Sex: steer, −6; bull, +14; heifer, −8.

(4) Per one percent inbreeding of calf: −1.75.

(5) Per one percent inbreeding of dam: −1.15.

(6) Per increase in age of calf of one day: +1.67.

Literature Cited

Black, W. H. and B. Knapp, Jr. 1936. A method of measuring performance in beef cattle. Amer. Soc. Anim. Prod. Proc. pp. 72–77.

Bloom, Philip E., 1953. Analysis of beef cattle production testing in Kittitas County, Washington, Proc. W. Sect. Amer. Soc. An. Prod. (4) II, 1–8.

Gannett, H. 1906. A dictionary of altitudes. Dept. of Int., U. S. Geol. Survey, Bul. 274, Series F, Geography, 47.

Hazel, L. N. 1946. The covariance of multiple classification tables with unequal subclass numbers. Biom. Bul. Z (2) 21–25.

Henderson, C. R. 1948. Estimation of general, specific, and maternal combining abilities in crosses among inbred lines of swine. Ph.D. dissertation. Iowa State College of Agr. and Mech. Arts, Ames, Ia.

Knapp, B. Jr., and W. H. Black. 1941. Factors influencing rate of gain of beef calves during the suckling period. Jour. Agr. Res. 63:249.

Knapp, B. Jr., A. L. Baker, J. R. Quesenberry and R. T. Clark. 1942. Growth and production factors in range cattle. Mont. Agr. Expt. Bul. 400.

Knox, J. H. and M. Koger. 1945. Effect of age on the weight and production of range cows. N. Mex. Agr. Expt. Sta. Press Bul. 1004.

Koch, Robert M. 1951. Size of calves at weaning as a permanent characteristic of range Hereford cows. JOURNAL OF ANIMAL SCIENCE 10:768.

Koger, M. and J. H. Knox. 1945. The effect of sex on the weaning weight of range calves. JOURNAL OF ANIAML SCIENCE 4:15.

Landblom, Nellie. 1954. Unpublished notes. Colorado Agr. Exp. Sta., Fort Collins, Colorado.

Lush, J. H. 1945. Animal breeding plans. 3rd ed. Iowa State College Press, Ames. Ia.
Rollins, W. C., H. R. Guilbert and P. W. Gregory. 1952. Genetic and environmental factors affecting preweaning growth of Hereford cattle. Am. Soc. An. Prod. West. Sect. Proc. 3:X:1.
Sawyer, W. A., R. Bogart and M. M. Oloufa. 1948. Weaning weight of calves as related to age of dam, sex, and color. JOURNAL OF ANIMAL SCIENCE 7:514. Abstract.
Yearbook of Agriculture. 1941. Climate and man. U.S.D.A., U. S. Govt. Printing Office, Washington, D. C.

THE INDIVIDUALITY OF THE LEVEL OF BLOOD GLUTATHIONE IN YOUNG BEEF CATTLE [1]

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THE concept that individual human beings and certain laboratory animals possess distinctive patterns of metabolic traits has been established by Williams *et al.* (1949). The hypothesis has been set forth that the metabolic patterns of individuals are of great importance with respect to susceptibility to disease, nutrition, mental abilities, sexual behavior, and many other facets of human life.

An extension of the above stated concept would be that animals of economic importance possess distinctive patterns of metabolic traits. Also, the metabolic pattern may be related to the ability of the animal to withstand adverse environmental conditions, to nutritional requirements and to productivity. With this possibility in mind, the study of metabolic or biochemical individuality becomes a matter of interest, particularly with respect to the heritability of individual differences.

Evidence suggesting that cattle possess a metabolic individuality has been presented in the case of two constituents of the blood serum: the protein bound iodine and the alkaline phosphatase. Breed differences have been demonstrated in the case of the protein bound iodine by Long *et al.* (1952) and of the phosphatase by Kunkel *et al.* (1953). Data are not yet available to show conclusively that these substances are largely unaffected by environmental factors.

It would be desirable in establishing experimental support of the concept of metabolic individuality in cattle that there be found at least one chemical constituent of a tissue, variations in the level of which are relatively unaffected by environmental changes but which is as much a characteristic of the individual animal as the color pattern of the hair coat or the shape of the head. In the present work, evidence is presented to indicate that the glutathione content of the blood of beef cattle 9–13 months of age, is an individual characteristic.

[1] This investigation was conducted in cooperation with the Livestock Division, Agricultural Research Service, United States Department of Agriculture, as a part of the Southern Regional Beef Cattle Breeding Project S–10. The cooperation of Mr. J. K. Riggs, Mr. T. C. Cartwright, Mr. David K. Stokes, Jr., and Dr. B. L. Warwick in obtaining the samples of blood is gratefully acknowledged.

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Materials and Methods

The subjects of this investigation were beef cattle, 9–13 months of age at the time the first samples were taken, in five record-of-performance feeding trials during the years 1951 to 1953. At the time of the test all animals were receiving an adequate ration for growth. In some cases, the test cattle had been brought in to the test site from various areas of Texas while in other cases cattle were tested at the site at which the parental herd was kept.

Glutathione was determined in whole blood by the method of Grunert and Philips (1951). Blood samples were collected by venepuncture into tubes containing potassium oxalate as the anticoagulant. All samples were cooled in an ice bath shortly after collection and were kept at about 0°C. until analyzed. All analyses for glutathione were accomplished within twelve hours after taking the samples. In preliminary tests, no significant loss of glutathione could be detected within this period of time. When used for comparative purpose, hemoglobin was determined colorimetrically as oxyhemoglobin.

Results and Discussion

Estimates of Repeatability

The correlations between determination of glutathione levels of the same animal obtained at different ages were calculated for five groups of Hereford and Angus cattle totaling 69 animals (table 1). In the case of the Brahmans studied, only one determination was made for each animal and therefore, estimates of repeatability were not calculated. The groupings for these calculations were based upon handling procedures, breed and test location. With the exception of group 1, where the intraclass correlation was used, the repeatabilities were calculated as the product-moment coefficients of correlation between two successive determinations from the same animal. Mean glutathione values and the calculated coefficients of variations are presented for each sampling date.

The estimates of repeatabilities for these five groups ranged from 0.74 to 0.97, correlations which were highly significant statistically. Thus, it appears that from 50% to 90% or more of the observed variation in the level of blood reduced glutathione is due to relatively permanent differences between the animals.

The average glutathione level remained relatively constant within groups during the observations made in this study. Reid, Ward and

Salsbury (1948) reported a gradual increase in blood glutathione with increasing age. The data obtained in the present study suggest a slight rise in the average glutathione level with the subsequent determinations, but the greatest change observed to occur in a six-month period was only a 13% increase in average glutathione. In addition, the cal-

TABLE 1. ESTIMATES OF REPEATABILITY OF BLOOD GLUTATHIONE LEVELS

Group No.	Group Description	No. of Animals	Date of Sampling	Av. Glutathione	Coefficient of variation	Repeatability
				mg./100 ml.	%	
1	Hereford bulls	10	Oct.	24.7	17.1	
			Dec.	27.2	11.6	
			Feb.	26.0	12.9	
			Apr.	28.0	10.1	0.78
2	Hereford heifers	12	Jan.	30.3	12.3	
			Apr.	30.4	14.6	0.74
3	Hereford bulls	8	Jan.	32.5	25.0	
			Apr.	34.4	19.4	0.93
4	Hereford steers and heifers	21	Oct. 14	23.6	22.4	
			Nov. 23	26.7	20.9	0.92
5	Angus steers and heifers	14	Oct. 14	33.2	49.0	
			Nov. 23	35.6	46.0	0.968

culated coefficients of correlation between age and blood glutathione at a given time ranged from −0.21 to 0.01 for the various groups of animals. Thus the observed differences in glutathione levels do not appear to be the result of differences in the age of the animal.

The calculated coefficient of variation also remained relatively constant within groups but varied widely between groups. In view of the fact that high correlation coefficients were obtained when the coefficients of variation were relatively high, the feeling is gained that errors in the chemical analyses accounted for most of the variation statistically not accounted for by animal differences.

Breed Comparisons

In the course of this study, it was possible to compare young Hereford cattle with Angus cattle of the same sex and of similar age in two tests and with Brahman cattle in another case. The observed ranges

with the average glutathione levels, standard error and coefficients of variation are given for each breed in table 2.

It is to be noted that the observed ranges of glutathione levels in the Angus were much wider than in the Herefords. The largest coefficient of variation calculated for a group of Herefords was 25% while in the Angus the smallest coefficient was 29%. In addition, in the bulls

TABLE 2. COMPARISON OF BLOOD GLUTATHIONE IN YOUNG HEREFORDS WITH THE ANGUS AND BRAHMAN BREEDS

Test No.	Sex	Breed	No. of animals	Glutathione, mg./100 ml. blood			Coefficient of Variation
				Range	Av.	S.E.	
							%
1	Bulls	Hereford	7	17–24	19.7	1.2	15.5
		Angus	13	22–53	27.6**	2.2	28.9
2	Steers	Hereford	12	18–30	26.8	1.5	19.8
		Angus	7	25–76	35.4	6.9	51.6
	Heifers	Hereford	10	15–30	23.7	1.4	18.1
		Angus	8	20–69	35.4	5.2	41.7
3	Heifers	Hereford	17	25–33	28.9	0.8	11.6
		Brahman	36	24–57	34.1**	1.1	18.7
	Bulls	Hereford	11	15–31	24.4	1.3	18.0
		Brahman	15	28–37	31.7**	0.9	11.3

** Values significantly ($p < 0.01$) greater than those of the Herefords.

(listed as test 1 in table 2) the average glutathione level in the Angus was significantly greater than in the Hereford. The difference between these breeds was not significant on a within-sex basis in the second comparison, but when the data from steers and heifers were combined, the breed difference was highly significant statistically. As far as the authors are aware, this is the first time a significant biochemical difference, other than hair coat pigment, has been demonstrated between these two beef breeds.

The levels of reduced glutathione in the blood of young Brahman cattle also were found to be significantly higher than in Herefords of similar age. The data obtained from the Brahman cattle, however, did not show the high coefficients of variations as did those from the Angus.

Estimate of Heritability

The breed differences in reduced glutathione level in the blood indicate that observed differences are contributed to by differences in heredity, at least, on a between-breed basis. The high repeatability of successive determinations of glutathione with breed groups would lead one to expect to find a high heritability within breeds since it

does not seem plausible that permanent environmental differences could be contributing much to the repeatability.

Sufficient data were not available to obtain an estimate of heritability within the Angus or Brahman breeds. Enough data were obtained to note significant sire differences when the average glutathione values were considered without regard to sex (table 3). This pooling of sexes was based on the fact that no significant differences were found between sexes within sire groups.

TABLE 3. ANALYSIS OF VARIANCE OF GLUTATHIONE LEVELS IN HEREFORD CALVES

Source of Variation	D/f	Mean Square	Theoretical Composition of Mean Square
Total	96		
Years	2	133.13*	
Between sire groups	11	30.98*	$I + kS$
Within sire groups	83	13.75	I

I = Variance due to differences between individual offspring of the same sire.
S = Variance due to differences between sires.
$k = \frac{97}{14} - \frac{V_n}{14} = 4.57$ where V_n is the variance of number of animals per sire group.
* Probability of less than 0.05.

The heritability estimated by the half-sib correlation method of Lush (1949) from the data in table 3 was 0.86. While this must be admitted to be a preliminary estimate, it is not an unreasonable one since estimates of repeatability as high as 0.93 were obtained with some of the groups of Herefords. It would thus appear that the level of reduced glutathione in the blood of young beef cattle is a highly heritable characteristic.

Discussion

Since the glutathione of the blood is found primarily in the erythrocyte, some of the variation in the level of this compound may be the result of differences in numbers of red cells in the blood. There appears to be some relationship between red cell concentration and glutathione level in the Hereford. In a group of 12 bulls and 18 heifers of the Hereford breed, the calculated coefficient of correlation between the reduced glutathione level and the hemoglobin content of the blood was 0.50, a correlation which was highly significant statistically. However, in a group (14 bulls, 36 heifers) of Brahman cattle the correla-

tion was a —0.10. Thus variation in number of red cells, as indicated by the hemoglobin concentration, may account for 25% of the variation in the level of blood glutathione in the Hereford, but it does not account for individual differences in the Brahman cattle. Although hemoglobin data were not obtained from the Angus cattle, it is very unlikely that blood cell or hemoglobin concentration could account for the 20 to 76 mg.-per-100 ml range in glutathione concentration observed in these animals.

Gregory and Goss (1933) suggested that glutathione may be an important biochemical substance involved in the regulation of body size in rabbits. In the present investigation, the data obtained thus far have not revealed any particular relationship of the glutathione level with the feedlot rate of gain or the efficiency of feed utilization in cattle.

Considerable time and space could be devoted to a discussion of the possible biochemical and physiological significance of the data obtained in the present work. However, in view of the high degree of speculation which would be required, it is perhaps best simply to point out that the reduced glutathione level in the blood of young beef cattle is a characteristic of each animal and as such provides evidence that biochemical individuality does indeed exist in cattle.

Summary

The reduced glutathione content of blood was determined in Hereford, Angus, and Brahman cattle 9–13 months of age. In the Hereford and the Angus breeds repeatability estimates ranging from 0.74 to 0.97 were obtained with subsequent determinations made one to six months apart.

The average level of blood glutathione was found to be significantly lower in the Hereford breed than in Angus or Brahman cattle.

A preliminary heritability estimate of 0.86 was obtained for the glutathione level in young Hereford cattle.

Twenty-five percent of the variation in glutathione level in the Herefords could be accounted for by variation in the hemoglobin concentration. No significant relationship of the level of glutathione with hemoglobin content of blood was noted in the Brahman cattle.

It is believed that these data provide definite and specific evidence of biochemical individuality in cattle.

Literature Cited

Gregory, P. W., and H. W. Goss. 1933. The relation of sullphydryl concentration to size inheritance in the rabbit. Amer. Naturalist 67:180.

Grunert, R. R., and P. H. Phillips. 1951. A modification of the nitroprusside method of analysis for glutathione. Arch. Biochem. 30:217.

Kunkel, H. O., D. K. Stokes, Jr., W. B. Anthony, and M. F. Futrell. 1953. Serum alkaline phosphatase activity in European and Brahman breeds of cattle and their crossbred types. JOURNAL OF ANIMAL SCIENCE 12:765.

Long, J. R., L. O. Gilmore, S. M. Curtis, and D. C. Rife. 1952. Bovine protein bound iodine and its relation to age and breed. Jour. Dairy Sci. 36:603.

Lush, J. L. 1949. Heretability of quantitative characters in farm animals. Proc. 8th International Congress Genetics. (Hereditas Suppl. Vol.) 356–375.

Reid, J. T., G. M. Ward and R. L. Salsbury. 1948. Blood glutathione in the bovine. Amer. Jour. Physiol. 152:633.

Williams, R. J., L. J. Berry and E. Beerstecher, Jr. 1949. Individual metabolic patterns, alcoholism, genetotrophic diseases. Proc. Nat. Acad. Sci. 35:365.

IMPAIRED REPRODUCTION IN THE RABBIT FED SUPPLEMENTED DIETS CONTAINING SOYBEAN HAY

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IMPAIRED reproduction as well as evidence of vascular breakdown has been reported in the rabbit fed diets containing soybean hay (Kendall *et al.* 1950). These abnormalities were not observed in what were considered normal New Zealand White female rabbits whose diets contained a roughage other than soybean hay namely: lespedeza, alfalfa or timothy hay. Litter sizes of females fed diets containing the latter roughages averaged 8.4 young per litter with no evidence of abnormal body function either in the females or in their young. This was in contrast to the performance of females in a series of studies fed a diet containing soybean hay, whose litter size was 5.8 young per pregnancy and was frequently accompanied by vascular breakdown in different parts of the body, as well as evidence of fetal mortality during the latter half of the gestation period.

In the earlier report (Kendall *et al.*, 1950) a number of questions were raised as to the possible cause or causes for the observed syndrome in the rabbit fed a soybean hay-wheat diet. A number of the questions raised grew out of the cited studies in which nutritional deficiencies have been reported when the soybean or its fractions have been fed to other animals. The close similarity of this syndrome to other cited syndromes in which impaired reproduction had been associated with certain vitamin deficiencies, was called to the reader's attention. Therefore, the purpose of this report is to summarize the results of a number of studies that were conducted with the idea of attempting to answer some of the questions raised earlier by testing the effects of administering certain supplements, namely: d-l methionine, β-carotene, vitamin A acetate, α-tocopherol, 2 methyl-naphthoquinone, vitamin B_{12}, choline, bonemeal and progesterone as well as the feeding of autoclaved soybean hay upon the possible alleviation of a sterility syndrome which occurs in the rabbit fed diets containing soybean hay.

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Experimental

Three series of experiments were conducted and are designated as experimental series 1, 2 and 3. In all three series, the basic diet fed consisted of mow-cured, finely ground, soybean hay 49.5 parts, mixed with finely ground wheat 49.5 parts and sodium chloride 1 part. The composition of this diet as fed as determined by proximate analysis (AOAC 1945) was dry matter 88.5 percent, total protein (nitrogen x 6.25) 15.1 percent, ether extract 3.1 percent, crude fiber 13.1 percent, nitrogen free extract 51.5 percent and ash 5.7 percent. The carotene content of the soybean hay as fed averaged approximately 20γ per gram of hay.

In planning these investigations it was considered unnecessary to include roughages other than soybean hay in the diets to serve as normal controls. Such an assumption was believed reasonable since the first positive evidence of impaired reproduction was observed in ten New Zealand White females whose diet contained soybean hay and whose litters averaged 3.2 young per mating. This same group of females had previously reproduced normally, when fed the same diet except that it contained lespedeza instead of soybean hay, with an average of 8.9 living young per mating. Further justification for omitting roughages other than soybean hay from the diets for control purposes was supported by the fact that when alfalfa replaced soybean hay in the diet the litter size for 14 females of similar breeding was 8.0 young per mating.

Two breeds of rabbits were used, namely, the New Zealand White and Dutch breeds whose average weights were 3.5 kg. and 2 kg. respectively. The animals were housed in individual cages and were fed *ad libitum*. Fresh water was available to the animals at all times. The animals were placed on the diet on the same day they were mated to fertile males.

Experimental series 1.

In this series a number of supplements were administered or fed to the mated females in addition to the above described basic soybean hay-wheat diet. Each supplement tested was fed or administered to five females with the exception of one group (vitamin B_{12} group) which included seven females. The following supplements were given at the indicated rates or were included in the diet at the indicated level: d-l methionine added to make up 0.3 percent of the diet; choline

chloride 0.3 percent; steamed bone meal 1.0 percent; carotene (90 percent β 10 percent α) in cottonseed oil administered orally each day, 200 τ per animal; vitamin A acetate in cottonseed oil administered orally each day, 200 τ per animal, α-tocopherol, orally administered, at the rate of 4 mg. per kg. body weight; progesterone, 2 mg., were injected subcutaneously per animal daily; 2 methyl-naphthoquinone, 2 mg. per animal daily; vitamin B_{12} 100 τ was given orally two times per week. The females in this series were permitted to kindle at 31 days.

During the course of gestation observations were made for any abnormal symptoms: hemorrhage, abortion, or voiding of partially resorbed fetuses.

Experimental series 2.

In this series of experiments, two levels of α-tocopherol were administered daily to mated females receiving the basic diet of soybean hay and wheat. In one experiment 10 mg. of α-tocopherol were given orally to one group of seven females, to another similar group 10 mg. of α-tocopherol were injected subcutaneously. Seven females receiving only the basic diet were used as controls. To another group of six females a level of 60 mg. of α-tocopherol were given orally each day. A similar group of six were used as controls in this experiment.

On the 28th day after mating the animals were sacrificed and autopsied. Observations were made of the number of living young; the number of resorption sites; the number of corpora lutea as well as any abnormalities which were evident.

In some experiments of this series only blood plasma samples were taken for vitamin A and tocopherol analysis. In other experiments blood plasma and liver samples were taken for vitamin A analysis. Vitamin A in blood plasma was determined by Kimble's (1939) method modified by using activated glycerol dichlorohydrin for developing the color (Sobel and Werbin, 1946). For determining liver vitamin A the methods of Gallup *et al.* (1946) and Sobel and Werbin (1946) were used. Vitamin A values were obtained through the use of a Coleman Spectrophotometer, Model 14. Blood plasma tocopherol was determined by the method of Quaife *et al.* (1944).

Experimental series 3.

This series was conducted for the purpose of studying the influence of heating the hay by autoclaving upon the alleviation of the sterility

symptoms. This was done with the idea that any possible enzymes, if present in the hay and were factors in the syndrome, would be inactivated through the action of heating. An attempt to accomplish this was made by autoclaving soybean hay for 30 minutes at 120°C. and at a pressure of 15 pounds. The autoclaved hay was used in compounding a diet of the same proportions as the basic diet. Five groups of five females each were used in this series. Groups 1 and 2 received a diet containing the autoclaved soybean hay. Groups 3, 4 and 5 received the basic diet. In addition groups 2 and 5 received 50 mcg. of β carotene daily per kg. of body weight. Group 4 received 80 mcg. of vitamin A.

On the 28th day following mating by sacrificing and autopsying the females, observations were made of the number of living young, the number of sites of resorption, the number of corpora lutea. Plasma and liver samples were taken for vitamin A analysis.

Results

In tables 1, 2 and 3 the reproduction performance of the mated females is presented.

Under the conditions of these studies d-l methionine failed to prevent the onset of hemorrhage. Two females out of five in this group homerrhaged severely. The number of young born for this group (table 1) was well below normal of the two breeds used in this study. Normal average litter sizes per mating for the Dutch and New Zealand White breeds of rabbits as observed in this laboratory have been approximately 6.1 and 8.3 young per mating respectively for the Dutch and New Zealand White breeds.

Although those females receiving carotene in this series failed to show evidence of hemorrhaging, the number of young born was below normal (table 1) as was also true when α-tocopherol was used as a supplement to the basic diet.

Progesterone when administered at the indicated levels, was ineffective in promoting normal litter size (table 1) even though no hemorrhage was observed in this group.

Further evidence was shown in this study (table 1) that the cause for hemorrhage was probably not due to a lack of vitamin K since in this group of five females, 2 methyl-naphthoquinone appeared to be without effect in controlling hemorrhage.

When the basic diet was supplemented with either choline or steamed bone meal, females continued to abort fetuses as well as to show evidence of high fetal mortality having occurred during the latter part

of the gestation period as observed at autopsy on the 28th day following mating.

The observations made in this series of studies indicate that the symptoms typical for the syndrome, namely, hemorrhage emanating from the reproduction tract, fetal resorption and abortion, are not specific for a single breed.

TABLE 1. THE REPRODUCTIVE PERFORMANCE OF FEMALE RABBITS AS ASSOCIATED WITH FEEDING A SUPPLEMENTED SOYBEAN HAY-WHEAT DIET

Supplement	Females			Young carried 28 days or to term			Abnormal symptoms observed during gestation
	D[1]	NZ[2]	Known pregnant	Living	Dead	Corpora	
				Series IA			
No supplement	4	1	5[3]	26	3	—	2 cases severe hemorrhage. 1 female died 22nd day of preg.
d–l methionine	4	1	4[3]	13	2	—	2 cases of severe hemorrhage
β-carotene	4	1	4[3]	25	2	—	None
α-tocopherol	4	1	3[3]	17	0	—	1 female voided tissue remnants 28th day
2 methyl-naphthoquinone	4	1	3[3]	20	3	—	1 abortion at 14 days followed by severe hemorrhage
Progesterone	4	1	4[3]	14	2	—	None
Vitamin B_{12}	0	7	5[3]	22	24	—	2 cases severe hemorrhage
				Series IB			
No supplement	0	5	4[4]	12	17	39	1 abortion 27th day
Choline	0	5	4[4]	10	25+	37	1 abortion 26th day
Steamed bone meal	0	5	4[4]	6	17	39	1 abortion 27th day

[1] Dutch breed.
[2] New Zealand White breed.
[3] 31 days after mating.
[4] Sacrificed 28 days after mating.

The results of the second series of experiments where higher levels of α-tocopherol than those which were administered in experimental series 1, indicate that the diet was not improved as indicated by the observed reproductive performance. The high mortality rate in the young observed on the 28th day after mating where 10 mg. of α-tocopherol was fed (table 2) suggests the possibility of a higher concentration of the syndrome including factor(s) in this particular lot of soybean hay. Feeding 60 mg. α-tocopherol daily per animal failed to promote an improved reproductive performance over that of the controls.

In the experimental series 3, where autoclaved soybean hay replaced the regularly used soybean hay, the results indicate that autoclaved

hay did not improve the reproductive performance over that observed where the non-autoclaved hay was included in the diet.

In many of the autopsy studies light colored placentae were observed. In some instances some fetuses in litters were considerably smaller than litter mates.

Discussion.

The evidence shows that the sterility syndrome does affect reproduction after implantation has occurred. The numbers of implantations per

TABLE 2. THE REPRODUCTIVE RESPONSE TO INCREASED LEVELS OF α-TOCOPHEROL SUPPLEMENTATION

α tocopherol Administered	Females mated	Females showing evidence of pregnancy: At 28 days	Living young	Dead young	Corpora	Vitamin A: Plasma γ/100 ml.	Liver γ/gram	Tocopherol plasma mg /100 ml.
None	13	11	53	35	73	38	24.4[2]	.495[3]
Injected (10 mg.)	7	7	0	24	18	29	——	.427
Oral (10 mg.)	7	4	12	20	39	26	——	.778
Oral (60 mg.)	6	5	31	8	47	60.3	38.3	——

[1] All females were of New Zealand White breed.
[2] Average liver vitamin A values for 6 animals.
[3] Average for 7 animals.

mating or per individual frequently have been equal to those of normal reproductive performance. The appearance of small fetuses in some instances and the presence of light colored placental membranes suggest a possible lowered vascularity within the placenta which may have resulted in a lowered nutrient supply thus affecting the developing fetuses to a varying degree. This raises the question as to whether the syndrome may be due in part to a partial failure in placental function which may involve some endocrine relationships. That threshold levels of impaired placental function may have existed are suggested by the presence of varying numbers of fetuses, some small and some normal size. In addition the presence of varying numbers of sites of resorption ranging in age from highly advanced stages of resorption to fetuses having died within 24 hours before the time of autopsy offers further evidence that possible threshold levels of impaired placental function may have occurred.

It is apparent that the syndrome was not prevented by supplementing the soybean hay-wheat diet at the indicated levels with d-l methionine,

carotene, α-tocopherol, vitamin A, 2 methyl-naphthoquinone, progesterone, vitamin B_{12}, choline or bone meal.

It appears that vitamin A levels in the plasma and liver as well as levels of vitamin E in plasma, though somewhat variable between individual animals, for the most part were normal. That the syndrome appears in animals having normal levels of these vitamins is evidence

TABLE 3. REPRODUCTIVE PERFORMANCE AS ASSOCIATED WITH AUTOCLAVED AND NON-AUTOCLAVED SOYBEAN HAY IN THE DIET

Dietary factor or factors	Females[1]		Females showing evidence of pregnancy at 28 days				Vitamin A	
	Mated	Aborting	Pregnancy positive	No. living young	Site of resorption	No. Corpora	Plasma γ/100 ml.	Liver γ/gram
Autoclaved hay	5	0	5	28	8	58	50.0	39.0
Autoclaved hay plus carotene	5	2[2]	5	10	22	47	51.0	34.2
Basic diet	5	1[2]	5	23	25	50	31.5	34.3
Basic diet plus vitamin A	5	0	5	30	14	55	44.4	40.5
Basic diet plus carotene	5	0	5	26	11	51	38.7	60.0

[1] One animal of the Dutch breed and 4 animals of the New Zealand White breeds were included in each group.

[2] One case of severe hemorrhage in each group.

which discounts the possibility of any reduction in vitamin levels in females due to carotene destruction in the hay curing process or in the gastrointestinal tract of the animals during digestion.

It does not seem likely that a lack of dietary provitamin A in the soybean hay fed rabbit would be a factor contributing to the syndrome's appearance. This assumption seems reasonable on the basis of evidence gained in other studies in this laboratory where normal reproduction has been observed in normal female rabbits when placed on a carotene deficient diet at mating time and thus fed without vitamin A supplementation throughout pregnancy.

The fact that the administration of progesterone at the rate of 2 mg. daily appeared to be ineffective in alleviating reproductive failure does not preclude the possibility that higher levels of progesterone administered may have an alleviating effect. The effects of administering higher levels of progesterone upon the syndrome's alleviation are now being studied.

The failure of the animals to carry their young to term is suggestive of impaired nutrition for normal growth of the fetus. This may be due

to abnormal physiological function of the placenta. Whether the presence of a toxic factor is contributing to the sterility condition is not evident from these studies. Studies are now in progress to test the effects of feeding certain fractions of the hay as they may relate to the syndrome in an attempt to isolate more definitely the factor or factors that may be contributing to the failure in reproduction.

Summary

The results of three series of experiments in which further study of the sterility syndrome in the rabbit fed soybean hay are reported.

Supplementing the soybean hay-wheat diet at the indicated levels with d-l methionine, carotene (β 90 percent and α 10 percent) α-tocopherol, 2 methyl-naphthoquinone, vitamin A, progesterone, vitamin B_{12}, choline or steamed bone meal failed to alleviate the sterility symptoms or improve the reproductive performance of females over that of females receiving no supplements.

Plasma and liver vitamin A levels in the females fed the soybean hay-wheat diet appeared to be normal without carotene or vitamin A supplementation. Plasma tocopherol levels also appeared normal. These findings suggest that this syndrome is not directly associated with the levels of these vitamins even though the nature and incidence of the syndrome parallels in many respects that of the vitamin A or E deficient animal.

The feeding of soybean hay, autoclaved at 120°C. for 30 minutes, was ineffective in alleviating the syndrome. This would suggest that the causative factor or factors are probably not enzymic in nature.

Literature Cited

Association of Official Agricultural Chemists. 1945. Official and Tentative Methods of Analysts. 6th ed., Washington, D. C.

Gallup, Wallis D. and J. A. Hoeffer. 1946. Determination of vitamin A in liver. Ind. Eng. Chem. Anal. Ed. 18:288.

Kendall, K. A., G. W. Salisbury and N. L. VanDemark. 1950. Sterility in the rabbit associated with soybean hay feeding. Jour. Nutr. 42:487.

Kimble, M. S. 1939. The photocolorimetric determination of vitamin A and carotene in human plasma. Jour. Lab. Clin. Med., 24:1055–1065.

Moore, L. A. and R. Ely. 1941. Extraction of carotene from plant material. Ind. Eng. Chem. Anal. Ed. 13:600.

Quaife, Mary Louise and P. L. Harris. 1944. The chemical estimation of tocopherols in blood plasma. Jour. Biol. Chem. 156:499.

Sobel, Albert E. and Harold Werbin. 1946. Activated glycerol dichlorohydrin a new colorimetric reagent for vitamin A. Ind. Eng. Chem. 18:570.

INCREASING SUMMER GAINS OF LIVESTOCK

With Cool Water, Concentrate Roughage, Wire Corrals, and Adequate Shades [1]

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WORK at the Imperial Valley Field Station has demonstrated four ways to greater livestock gains during hot months. The methods suggested are as follows: 1. Cool drinking water (Ittner, Kelly, and Guilbert, 1951) increased the daily gain of Hereford steers from 1.07 to 1.46 lb. 2. A concentrate-roughage diet has a lower increment of heat than does a roughage diet, permitting greater daily gains. 3. Wire pens and surrounding green vegetation assist daily gains, by subjecting the animal to less radiant heat than that given off by heavy corral fences and surrounding objects. 4. Shades (Ittner and Kelly, 1948, 1951) definitely reduce the heat load on animals.

This paper discusses studies in 1952–1953 of shades and cool drinking water additional to the studies cited above, together with investigations during the same period on the effects of ration and corral types and location on the comfort and consequent gains of livestock during hot weather.

Test Conditions

The corral pens in which tests were conducted were 50 x 60 ft., enclosed by six-foot fences constructed of four 2 x 10 in. planks. Each pen had a hay or aluminum shade 10 ft. above ground. The water-measuring devices, cool water tanks, water evaporation, water temperature recordings, and weather station operation were the same as those previously reported by Ittner *et al.* (1951).

Weather data for the summers of 1952 and 1953 are presented in table 1. Recorded temperature extremes were 113 degrees F. and 57 degrees F. in 1952, and 114 degrees F. and 63 degrees F. in 1953.

[1] This paper is a result of a cooperative project between the U. S. Department of Agriculture and the Divisions of Animal Husbandry and Agricultural Engineering of the University of California. The authors wish to acknowledge the assistance of Phil Trask and Delbert Gaskin, Herdsmen, and of W. C. Rollins, Assistant Professor of Animal Husbandry, for his work on the statistical analyses.

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Temperatures fell below 70 degrees F. on only five nights of 1952 and on twelve nights in 1953. Mean temperatures for the two years were almost the same (91.8 and 91.3 degrees F., respectively) and mean relative humidity was 40 and 36 percent, respectively. During both summers, thunder showers were fewer than normal.

TABLE 1. WEATHER DATA FOR THE EXPERIMENTAL PERIODS OF 1952 AND 1953

Period of Experiment	Max.	Min.	Mean	Mean [1] Relative Humidity	Noon Relative Humidity
1952	Degrees F.	Degrees F.	Degrees F.	%	%
July 2–July 29	106.8	76.0	91.4	40	26
July 30–Aug. 26	106.4	79.1	92.8	45	29
Aug. 27–Sept. 11	106.8	75.2	91.0	32	21
July 2–Sept. 11	106.7	77.0	91.8	40	26
1953					
June 30–July 27	106.7	80.8	93.7	38	27
July 28–Aug. 24	106.5	78.1	92.3	36	25
Aug. 25–Sept. 22	103.3	72.4	87.8	33	19
June 30–Sept. 22	105.5	77.1	91.3	36	24

[1] Every two hours the relative humidity was recorded throughout the experiment, then averaged to get these figures.

Test Results

Cool Drinking Water

In 1952 three lots of Hereford steers, consisting of 10 head per lot and averaging 768 lb. per head, received the following treatments during a 71-day test period, July 2 to September 11: lot 1, uncooled water; lot 2, cold water (65 degrees F.); and lot 3, cold water and a shower from 18 misty spray heads covering two-thirds of the corral area. All three groups were fed good quality alfalfa hay.

Cool drinking water tests in 1953 were combined with the ration experiments (see below). For these tests, four lots of seven Hereford steers averaging 871 lb. received the following treatments during an 84-day test period, June 30 to September 22: lot 1, cold water and hay; lot 2, cold water and hay plus grain; lot 3, uncooled water and hay plus grain; and lot 4, uncooled water and hay.

All groups during both years were fed twice daily, all the feed they would clean up between feedings. Every 28 days, the animals were weighed after a 10-hour stand in a dry corral without feed or water.

Uncooled drinking water for 1952 and 1953 averaged 89.6 degrees F. and 89.2 degrees F., ranging from 70 to 102 degrees F. and 72 to 104 degrees F., respectively. The daily temperature fluctuation was the same as previously reported (Ittner, Kelly, and Guilbert 1951). For both years the cooled drinking water was cooled to 65 ± 2 degrees F. with a large mechanical refrigeration unit (figure 1).

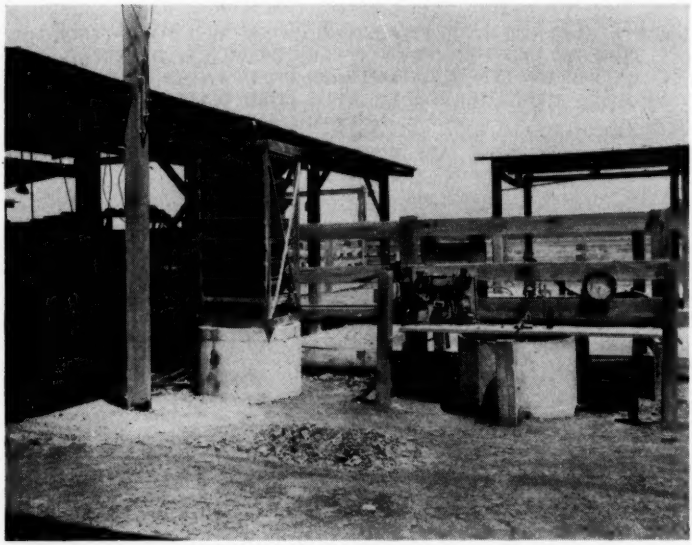

Figure 1. The mechanical refrigeration unit and equipment used for cooling the water to 65 ± 2 degrees F.

Rates of gain, food consumption, and feed per 100 lb. of gain for each group of animals are shown in table 2 (1952 test) and table 3 (1953 test). In 1952 the 10 Hereford steers on cold water gained 1.04 lb. per head per day, the warm water lot gained 0.78 lb, and the cold water-shower group made gains of 0.94 lb. The average daily gains for the 1953 tests were: lot 1 (cold water-hay), 1.51 lb.; lot 2 (cold water-hay plus grain), 2.01 lb.; lot 3 (warm water-hay plus grain), 1.57 lb.; and lot 4 (warm water-hay), 1.18 lb.

Average daily water consumption per animal, and per 100 lb. live weight, are given in table 4. In 1950, 1952, and 1953 tests, cold water groups drank less than animals receiving warm water. There was no meter on the drinking water tank of the warm water-hay group in

1953, but in 1950 and 1952 their water consumptions were almost the same—1.89 and 1.88 gallons per 100 lb. body weight respectively. There was some variation in water consumed by the cold water-hay groups during the three years, 1950, 1952, and 1953—1.69, 1.68, and 1.46 gallons, respectively, per 100 lb. body weight. The steers fed hay and grain in the 1953 tests drank less water than any other lot, lot 2 drank 1.17 gallons, and lot 3, 1.38 gallons per 100 lb. live weight.

TABLE 2. AVERAGE WEIGHTS, DAILY GAINS, AND FOOD CONSUMPTION OF TEN HEREFORD STEERS ON WARM DRINKING WATER, TEN HEREFORDS ON COLD WATER, AND TEN HEREFORDS WITH COLD WATER AND A MISTY SHOWER

(All figures are in lb.)

July 2–Sept. 11, 1952

Item	Lot 1 Warm Water	Lot 2 Cold Water	Lot 3 Cold Water Misty Showers
Av. initial weight	768	761	765
Av. final weight	823	835	832
Av. daily gain	0.78	1.04	0.94
Av. daily feed			
Alfalfa hay	19.36	20.35	20.80
Feed per 100 lb. gain			
Alfalfa hay	2481	1958	2221

Water consumption of cattle and the effect of cool drinking water are discussed at length in the 1951 report (Ittner, Kelly, and Guilbert).

Although the difference in daily gain in 1952, between the cold and warm water lots, was 0.26 lb. in favor of cold water, it was not significant. Ten animals per pen in 1952 may have been too many, especially in lot 2 (cold water) which had only 41 square ft. of shade per animal. These shades might have been adequate if some animals had not had to stand near the hot fence. Individual data also indicate that the "boss" animals may have selected the cooler spots for themselves.

The depression of daily gains in lot 3 (cold water, hay, and misty showers, 1952) may be explained by Kibler and Brody's (1953) recent studies on humidity effects. They showed that high humidity accompanied by high temperatures (75 to 100 degrees F.) caused a decided increase in rectal temperature and respiration rates of dairy cattle. The humidity may have been significantly increased, for the animals were under the misty sprays during most of the days.

A test in 1950 and two experiments in 1953 show that cool water (65±2 degrees F.) groups gain significantly faster than steers receiving warm water averaging 89 degrees F. Analysis of the 1953 studies show no significant interaction between feed and water effects; also, each of the main effects—feed and water—are significant at the 1% level.

TABLE 3. AVERAGE WEIGHTS, DAILY GAINS, AND FOOD CONSUMPTION OF HEREFORD STEERS ON COLD AND WARM WATER WITH DIFFERENT DIETS

(All figures are in lb.)

June 30–Sept. 22, 1953

Item	Lot 1 [1] Cold Water Hay	Lot 2 Cold Water Hay & Grain	Lot 3 Warm Water Hay & Grain	Lot 4 Warm Water Hay
Av. initial weight	867	881	869	868
Av. final weight	993	1049	1000	967
Av. daily gain	1.51	2.01	1.57	1.18
Av. daily feed				
Alfalfa hay	18.9	10.8	11.5	17.9
Oat hay	2.0	2.0	2.0	2.0
Barley and milo	..	5.8	4.3	..
Beet pulp	..	2.9	2.2	..
Total	20.9	21.5	20.0	19.9
Feed per 100 lb. gain				
Alfalfa hay	1259	539	737	1524
Oat hay	136	97	127	170
Barley & milo	..	291	277	..
Beet pulp	..	146	138	..
Total	1395	1073	1279	1694

[1] All lots started with 7 steers but one died in lot 2 from causes not associated with the experiment.

Effect of Ration

Feeding animals straw in winter to keep them warm through the increment of heat is well known, and by the same token a roughage diet during the hot summer can generate a lot of waste heat, which interferes with beef production. The 1953 experiments show this conclusively.

As mentioned above, tests in 1953 of the effect of the rations were combined with cool drinking water tests. Of four lots of seven Hereford steers, two were fed a ration of hay plus concentrate; the other two received hay only. One lot of each group also received cool drinking

water. The roughage was good quality alfalfa hay and oat hay, while the concentrates were barley and milo fed at a 70:30 ratio, and molasses dried beet pulp.

TABLE 4. AVERAGE DAILY WATER CONSUMPTION PER ANIMAL AND AVERAGE DAILY WATER CONSUMPTION PER 100 LB. OF LIVE WEIGHT

(All figures in gallons consumed daily)

	Lot 1		Lot 2		Lot 3	
	Warm Water		Cold Water		Cold Water and Showers	
1952 Period of Experiment	Per Head	Per 100 lb. Weight	Per Head	Per 100 lb. Weight	Per Head	Per 100 lb. Weight
First, July 2–July 29	14.5	1.88	13.3	1.72	14.2	1.83
Second, July 30–Aug. 26	15.0	1.90	13.2	1.64	13.8	1.73
Third, Aug. 27–Sept. 11	15.8	1.94	13.8	1.67	14.0	1.71
Average: July 2–Sept. 11	15.0	1.88	13.4	1.68	14.0	1.75
	Lot 1		Lot 2		Lot 3	
	Cold Water and hay		Cold Water, hay and grain		Warm Water, Hay and Grain	
1953 Period of Experiment	Per Head	Per 100 lb. Weight	Per Head	Per 100 lb. Weight	Per Head	Per 100 lb. Weight
First, June 30–July 27	12.6	1.42	11.6	1.27	13.3	1.49
Second, July 28–Aug. 24	14.2	1.54	10.5	1.09	12.9	1.40
Third, Aug. 25–Sept. 22	14.0	1.45	11.6	1.14	12.6	1.30
Average: June 30–Sept. 22	13.6	1.46	11.3	1.17	13.0	1.38

As shown in table 3, with cold water available, lot 2, fed a hay-concentrate ration, gained 0.5 lb. per day more than lot 1, on a straight hay diet. With uncooled water (lots 3 and 4), the difference was 0.39 lb. For every ration tested, feed consumption fell off during the summer and it took more feed to make a 100 lb. gain; but a hay-concentrate ration reduced the increment of heat. However, it must be remembered that the caloric intake on a concentrate-hay ration can be so adjusted that there is more heat from a grain ration than from hay alone. Animals on a high calorie grain diet usually make their own adjustments on especially hot days.

As mentioned above, analysis of the 1953 studies shows there is no significant interaction between feed and water effects; and each of the effects (water and feed) were significant at the 1% level.

Fence Materials and Pen Location

Radiant heat from heavy corral fences in a small area seems to be enough to make a difference in the daily gain of beef cattle. During the 1953 tests, one lot of animals (lot 4, warm water-hay) was moved to a wire pen at some distance from the main corrals, as a matter of convenience, before it was realized that the difference in environment would be enough to affect gains. During this month (July) these animals made average daily gains of 1.76 lb. in the wire pen but during the next month lost a little or remained the same when fed at the main corral. The month of September was cooler and all lots made good daily gains.

Five other animals, not included in this 1953 test, were fed hay and grain with warm water at the main corral and it was difficult to keep them on feed; as soon as they were moved to the wire pen (July 28 to August 24) their food consumption increased, each consuming 11.2 lb. of concentrates per day, compared to only 9.3 and 7.1 pounds for lots 2 and 3 during the same period. Two groups of Hereford steers were fed out in this wire pen during the summers of 1951 and 1952. Gains were 2.42 lb. for some rather thin steers averaging 800 lb., and 1.79 lb. for some rather fleshy 1000 lb. steers. The dates of these tests were July 2 to September 12, and July 10 to September 3.

In August and September, while lot 4 was with the other lots in the main corral, environments in the wire pen and the main corral were compared, by radiometer, thermocouple, and black globe thermometer readings (figure 2). Use of these instruments is discussed by Kelly, Bond, and Lorenzen (1949). The instrument readings indicated a comfort advantage for the wire pen due to differences in air temperature and velocity, and to differences in radiation from surrounding surfaces.

The air temperature at the wire pens was usually 1.5 to 2 degrees F. lower, on the days readings were taken, and at times as much as 6 degrees F. lower than at the wooden corral. The air velocity was always greater at the wire pens. The black globe thermometer readings indicated heat loads as much as 10 Btu. per hr. (per square ft. of animal surface) less at the wire pens, benefiting the animals about as

much as lowering the air temperature 4 degrees F.[5] Since the air temperature was lower at the wire pens by about 2 degrees F. to start with, and the radiation heat load was less, the animals here were in an environment that was in effect 5.5 to 6 degrees F. less.

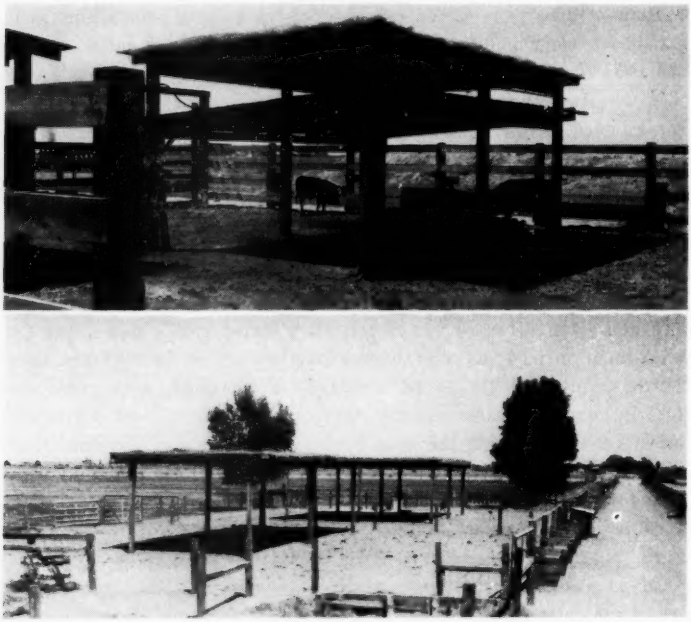

Figure 2. The upper picture shows the construction of the main corral while the lower one shows the wire pen used in these environmental studies.

Even though the comparisons between heavy corral fences and wire enclosed pens are not as direct as they might be, all the evidence shows that heavy fences radiate an excessive amount of heat. This extra heat causes a decrease in daily animal weight gain, a reduction in food consumption, and some difficulty in keeping feed consumption high enough for good gains. These corrals are much smaller and heavier than most commercial feed lot pens, and probably over-emphasize the radiant heat load from the fences. It was found in previous studies

[5] Based on comfort equation of Raber and Hutchinson (1947) i.e., environmental temperature is equal to one-half the sum of the ambient and mean radiant temperatures. For the level of mean radiant temperature considered here, 10 Btu per hr ft^2 represents a difference in mean radiant temperature of about 8 degrees F. as determined by black globe thermometer readings.

of shades (Ittner and Kelly, 1948 and 1951) that two environmental factors—air temperature and radiation—greatly affect animal gains. It was also shown that animals under shades whose roofs were cooled with water made better gains than those under shades with uncooled roofs. The effect of the cooler shade material is a reduction in radiant heat load. The effect of wire pens in place of wooden corrals is to combine two benefits—lower air temperature and less radiant heat load.

TABLE 5. SHADE PER ANIMAL IN SQUARE FEET DURING THREE SUMMER TESTS

Date					
Number of Animals	Shade in Sq. Ft.	Pen 1 627	Pen 2 640	Pen 3 588	Pen 4 408
	Treatment	Cold water alfalfa hay	Warm water alfalfa hay	....	....
1950	Av. daily gain lb.	1.46	1.07		
5	Sq. ft. of shade per animal	125	128		
	Treatment	Warm water alfalfa hay		Cold water shower alfalfa hay	Cold water alfalfa hay
1952	Av. daily gain lb.	0.78	....	0.94	1.04
10	Sq ft. of shade per animal	63	....	59	41
	Treatment		Warm water hay & grain	Cold Water hay & grain	Cold water alfalfa hay
1953	Av. daily gain lb.		1.57	2.01	1.51
7	Sq. ft of shade per animal		91	84	58

Each of four pens at the main corral had a hay or aluminum covered shade 10 ft. above the ground. These shades were thought to be adequate, but a summary of the data for 1950, 1952, and 1953 indicated at least one pen may not have had sufficient shade. The square feet of shade available to each animal for the three summer tests are summarized in table 5. Each pen had the same size and height of shade, but pens 2 and 3 received shade from the large central shade covering the scales and squeeze; pen 1 had some help from this shade, and from another small shade, but pen 4 had only the one shade. In 1950 the shade was more than adequate, while in 1952 lot 2 (in pen 4) would seem to have had an inadequate amount of shade—only 41 square ft. per animal. The next year, with only seven animals per pen, the shade area per animal was much better.

Although the amount of shade required by an animal has not been definitely determined, it is an important quantity and may vary with

the radiant heat load, which in turn will vary with size of corral, type of fence, and surrounding materials (vegetation or bare ground). At present it is felt that 60 square ft. of shade per animal is needed for cattle. An earlier discussion of shades was given by Kelly, Bond, and Ittner (1950).

Summary

Methods of keeping livestock cool during the summer in the Imperial Valley of California were investigated in 1952 and 1953. The mean air temperature during the test periods was 91.8 degrees F. for the 1952 period, and 91.3 degrees F. for 1953. The temperature of the cool drinking water was 65 ± 2 degrees F. for both years and the uncooled water averaged slightly over 89 degrees F. Work thus far shows that there are four ways of helping cattle keep cool and make faster gains during the summer.

1. Shades made of an insulating material like hay, or a reflecting material like aluminum, are best for reducing the heat load on the animals. For best results shades should be 10 to 12 ft. above ground. Adequate shade for each animal is important, and at present it is felt that 60 square ft. of shade per animal is needed for cattle.

2. Three years of experiments show that the steers that have access to cool drinking water will have a daily gain of from 0.26 to 0.44 lb. more than those on uncooled water. Three of the four tests completed were significant at the 1% level.

3. A roughage-concentrate ration for cattle produces less increment of heat, and faster gains, than a roughage diet. Hereford steers on cold water and alfalfa hay gained 1.51 lb. per head per day, while those on cold water-hay plus grain made 2.01 lb. On warm water the hay group gained 1.18 lb. while the hay and grain lot gained 1.57 lb. These differences are statistically significant at the 1 percent level.

4. Small heavy wooden corrals reflect and radiate enough heat to the steers to reduce their daily gains below those of animals in wire pens with some surrounding green vegetation. Measurements indicate an effective difference in environmental temperature equal to 5.5 to 6 degrees F.

Work at this station has definitely shown that proper precautions will materially increase the daily gains of animals during the summer. This aid in some cases at first seems to be so insignificant as not to be worth the effort, but it does actually tip the animals heat balance toward the zone of thermal neutrality, as the daily gains show.

Literature Cited

Ittner, N. R. and C. F. Kelly. 1951. Cattle shades. JOURNAL OF ANIMAL SCIENCE. 10:184.

Ittner, N. R., C. F. Kelly and H. R. Guilbert. 1951. Water consumption of Hereford and Brahman cattle and the effect of cooled drinking water in a hot climate. JOURNAL OF ANIMAL SCIENCE. 10:742.

Kelly, C. F., T. E. Bond and C. Lorenzen, Jr. 1949. Instrumentation for animal shelter research. Agricultural Engineering. 30:297.

Kelly, C. F., T. E. Bond and N. R. Ittner. 1950. Thermal design of livestock shades. Agr. Eng. 31:601.

Kelly, C. F. and N. R. Ittner. 1948. Artificial shades for livestock in hot climates. Agr. Eng. 29:239.

Kibler, H. H. and Samuel Brody. 1953. Environmental physiology and shelter engineering. Missouri Agr. Exp. Sta. Res. Bul. 522.

Raber, B. F. and F. W. Hutchinson. 1947. Panel Heating and Cooling Analysis. New York. John Wiley and Sons.

AN ESOPHAGEAL FISTULA FOR ANIMAL NUTRITION STUDIES

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VARIOUS methods have been used to make collections for determination of chemical composition and palatability of forage. For the purpose of chemical analyses, clipping has been used most extensively. This method gives not only the chemical composition of the plants but the yield as well. However, because of animal preference for some plants over others and their habit of grazing the plants at random heights, this method does not give an accurate picture of forage actually consumed by the animals, especially on pastures with a variety of forage plants. Other workers have taken forage samples by observing grazing animals and hand picking forage which they believe to be comparable to that consumed by the animals. This method is tedious and time consuming and the forage samples only approximate the feed actually eaten.

In order to obtain accurate samples for any of these determinations, it was thought desirable to use the animal itself as the collecting agent. To accomplish this, a series of trials was conducted with sheep which eventually resulted in the preparation of an esophageal fistula which would enable the animal to eat and drink normally but which could be opened allowing the forage eaten to be diverted through the fistula into a collection bag. In this way, it is possible to collect the fresh forage before it has undergone any chemical changes in the digestive system other than those occurring in the mouth.

Method

Three preliminary trials were conducted before a successful procedure was developed.

In the first trial, the method used by Pavlov (1910) in his work with the gastric juices of dogs was followed in which the esophagus was severed and the animal was fed through a rumen fistula. However, this necessitated hand feeding twice daily and it was difficult to keep the ewe on feed. Also it was thought that the fact that she was being fed ground

[1] The author wishes to acknowledge the help of Dr. A. C. McChesney, D.V.M., of Ukiah, California, who performed the surgery and offered technical advice concerning the operation involved.

alfalfa through the rumen fistula might affect her preference in forage. Rumen impaction resulted in the ewe's death.

In a second trial, an esophageal fistula was used. An effective means for closing the opening had not yet been developed. Bandages were tied around the neck by the use of elastic bands and were covered with a stockenette. When the sheep swallowed, the peristaltic movements traveled to the fistula with just a small movement carrying on down to the rumen, thereby forcing the feed out through the bandages. This resulted in malnutrition and eventual death to the animal.

Stainless steel pins were used as the means for closing the fistula in a third trial, allowing the esophagus to function normally when the animal was not being used for collection purposes. Since the pins had to be inserted at the time the fistula operation was performed in order to insure an adequate diet for the sheep, it was necessary to prevent the edges of the fistula from healing together by interposing gauze until union of the esophagus to skin was complete which required approximately ten days. With the increasing development of scar tissue, the fistula decreased in size to such an extent that grazed forage no longer dropped out. Surgery to enlarge the opening was unsuccessful and the sheep died fifty days after the first esophageal fistula operation was performed and ten days after the final operation.

In the fourth trial, results of which are reported herein, a two-year-old wether was selected and operated on as in trial 3. (figure 1.)

The method of choice for preparing esophageal fistulas of this type, as performed on this two-year-old wether, is as follows: An incision approximately one and one-half inches in length is made through the skin on the left side of the ventral surface of the neck and at a point midway between the jaw and the brisket. A stomach tube is then passed through the mouth and down the esophagus thereby affording a solid backing on which to make a similar incision in the esophagus with care being exercised to avoid cutting into the larger blood vessels supplying that area. The esophageal and skin incision edges are then approximated with interrupted nylon sutures inserted at close intervals, thereby resulting in fusion of the esophagus to the skin.

Two four-inch stainless steel pins are then inserted through the skin on either side of the opening parallel with the neck. These are entered at a point approximately one inch above and three-fourths inch on either side of the opening and passed through the subcutaneous fascia to a similar point at the lower end of the fistula. Permanent channels for easy insertion and removal of the pins are provided by passing Polyethy-

lene medical tubing over the entire length of the pins at the time of insertion. To prevent the pins from migrating toward the incision so rapidly and pulling out, thereby causing scar tissue, the tubing is sutured to the skin at about ⅜-inch intervals. By looping a cord over the ends of the pins in a lacing manner, the fistula can be closed and held in this position when the animal is not being used to make collections. During collection periods, the pins are removed so that the fistula remains open, allowing the forage taken by the animal to drop through the opening and into a receptacle which has been secured to the sheep.

Several methods of closing the fistula by use of the pins were tried. Rubber bands exerted a continual pressure on the skin and eventually pulled the pins through the skin. This was accomplished in only two days' time. The torn skin through which the pins had pulled healed very readily and formed scar tissue. The pins were reinserted further from the fistula and then pulled together by the use of a cord until the edges of the opening just barely touched.

As there is always some pressure on the pins when the opening is closed, necrosis takes place and the pins migrate toward the opening. However, this occurs quite slowly and the pins and medical polyethylene tubing can be reinserted. Some infection occurs at the insertion points but this can be minimized by the injection of penicillin ointment at the point of entrance.

Since the operation was made between two unsterile media with a sterile one between, sulfa salve was applied daily to the wound and daily injections of penicillin were administered for approximately a week. Very little infection was noted.

A bag constructed of canvas and lined with plastic to make it waterproof is secured to the sheep for collection purposes (figure 2). The posterior part of the bag is secured to the sheep by a strap around the heart girth with short straps running from the bag to the heart-girth strap. The bag is then secured about the neck by two straps which fasten at the top of the neck. A piece of stiffening between the two straps will hold the bag forward so that the container will always be under the fistula.

Results and Discussion

A series of four trials was conducted to prepare an esophageal fistula which would operate efficiently in making collections of forage consumed by sheep. In the final trial, a fistula was developed on a two-year-old wether which was fairly successful for this purpose. He had

been grazing on a range consisting of green annual forage and was supplemented with alfalfa hay. After the operation, he remained on the same feeding regime. Before initiating any specific studies, several

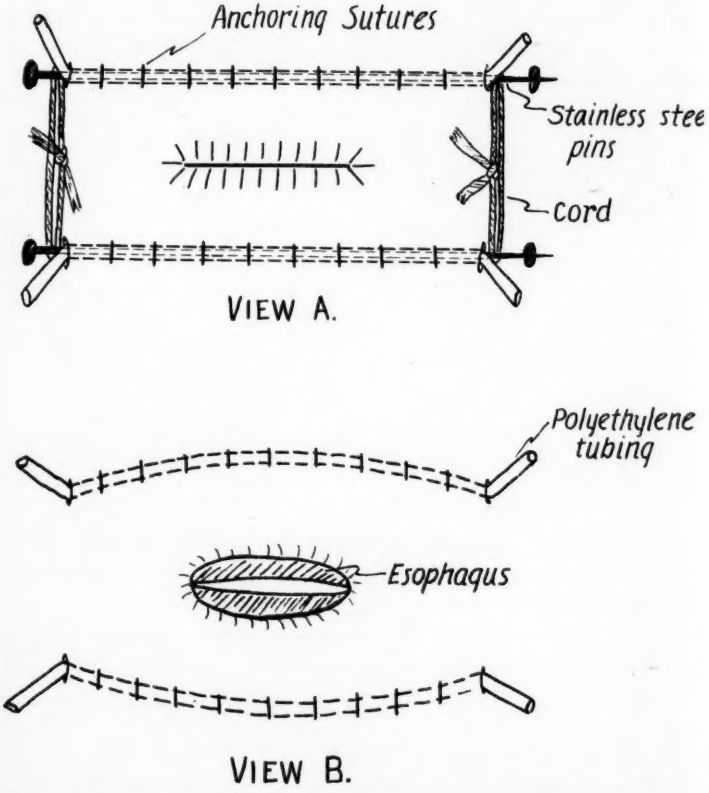

Figure 1. Sketches of esophageal fistula. View A—closed, View B—open.

preliminary trials were run to test this method of making such collections. Results of these trials are shown in table 1.

The percentage of forage which drops out through the fistula depends upon the size of the opening which has a tendency to become smaller with time. The desired size of the fistula may be maintained by periodical manipulation with the two forefingers. If the experiment underway requires that most or all of the forage eaten be collected,

then a larger opening should be prepared accordingly. The insertion of a cotton ball in the esophagus to insure collection of all of the grazed feed was not successful as an excessive amount of saliva was produced and the animal showed discomfort and would not eat.

So that samples will remain free of regurgitated material, collection periods should be of relatively short duration. If the animal is kept in dry lot overnight, it is likely that he will graze for several hours without regurgitating, especially if the weather is cool. The experimental animal should be kept away from water during collection periods because water running through the collected forage will leach out some of the nutrients.

TABLE 1. COLLECTION DATA

Date of Collection	Interval	Weight of Green Feed Collected	Air-Dry Weight
	Hours	Grams	Grams
2–18–54	3	298	34.9
2–18–54	5	699	67.5
2–22–54	6	1163	119.0
3–12–54	1	241	19.9

During the time when the animal is not being used for experimental purposes, the fistula is closed and eating and drinking habits appear normal. Occasionally there is some leakage upon regurgitation; however, this happens infrequently and the amount of feed lost through leakage is negligible.

Summary

An esophageal fistula has been devised for collecting samples of feed eaten by grazing animals. A series of trials is described in which a sheep with an esophageal fistula was used as the sampling and collecting agent for range herbage.

Addendum

An even more effective means of closing the fistula was developed in later trials. This method involved the fashioning of a plug from a plastic reagent bottle. Two discs were cut from the reagent bottle,

one approximately 3½ inches long and an inch wide, the other approximately 4 inches long and 2 inches wide. The two discs were

Figure 2. Bag attached to wether ready for forage collection

laced together with 60-pound braided nylon line, with the ends of the lace hanging loose through the larger of the two discs. When the animal is not being used to make collections, the smaller disc is forced through the fistula with the convex side toward the opening so that it

covers the inside of the fistula. The larger disc remains on the outside of the fistula with the convex side toward the opening and the nylon line is pulled taut, drawing the disc up against the outer skin. The lace is then tied in position. This type of plug, when in place, allows no leakage through the fistula.

Literature Cited

Pavlov, I. P. 1910. The work of the digestive glands. Translation by W. H. Thompson, Ed. 2, C. Griffin and Co. Ltd. 266 pp.

VITAMIN, AMINO ACID, AND ANTIBIOTIC SUPPLEMENTATION OF CORN-MEAT BY-PRODUCT RATIONS FOR SWINE

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MEAT by-products have played an important role in the nutrition of farm animals by balancing the deficiencies that exist in corn. As early as 1919 Hart and Steenbock revealed that meat proteins corrected the deficiencies of grains. Vestal (1946) observed that a ration of corn, tankage, alfalfa meal and minerals supported good growth of swine in dry lot. Subsequent research by Catron and Culbertson (1949) indicated crude vitamin B_{12} and APF concentrates could supply vitamin B_{12} and other unidentified growth factors as efficiently as animal proteins. Anderson and Hogan (1949) and Burnside *et al.* (1949) suggested that the addition of crude vitamin B_{12} concentrates and APF supplements to rations containing animal proteins stimulates growth.

More recently Lepley and associates (1950) observed that the addition of APF stimulated growth beyond the response obtained by adding meat and bone scraps to a corn-soybean oil meal ration. Luecke *et al.* (1951) reported the addition of streptomycin to a corn-soybean oil meal and to a mixed ration containing meat scraps resulted in a small increase in growth rate. Noland *et al.* (1951) found that pigs fed a ration containing meat scraps and an aureomycin-vitamin B_{12} supplement gained somewhat faster during the period from weaning to 100 lb. than pigs that were fed the unsupplemented meat scraps ration. In 1952 Briggs and Beeson reported the addition of vitamin B_{12} and/or antibiotics to plant protein rations resulted in a growth stimulation. These same authors had previously reported (1951) that the simultaneous addition of riboflavin, calcium pantothenate, and niacin to a basal ration containing both animal and plant proteins increased rate of gain and feed efficiency. Luecke *et al.* (1951) observed that the addition of liberal quantities of these same B-complex vitamins

[1] The authors wish to express their appreciation to Kingan and Company, Indianapolis, Indiana, for grants of both money and supplies and to the Dow Chemical Company for supplying the necessary amino acids through the courtesy of Dr. R. W. Colby.
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plus an antibiotic significantly improved a mixed animal and plant protein basal ration.

Research by Eggert *et al.* (1951), Blight *et al.* (1951), and Bloss *et al.* (1953) suggests the possibility of amino acid limitations in practical swine rations.

In light of such previous investigations with plant and combinations of plant and animal proteins, it was desirable to evaluate such supplementary effects on corn-meat by-product rations.

Trial 1

Experimental Procedure

The values of adding three B-vitamins (riboflavin, calcium pantothenate, and niacin) and an antibiotic-vitamin B_{12} supplement [3] to a protein supplement were studied under dry lot conditions. Cracked yellow corn, a mineral mixture (40 percent limestone, 40 percent steamed bonemeal, and 20 percent iodized salt), and protein supplements (table 1) were self-fed free choice. Thirty Duroc weanling pigs were divided into three lots of 5 barrows and 5 gilts per lot. Each group remained on trial until the pigs had attained an average weight of 200 lb.

Results

The results of Experiment 1 are presented in table 2. In this trial a basal ration of corn, tankage, alfalfa meal, and mineral was compared to similar rations which contained the three B-vitamins, terramycin, and vitamin B_{12}. Gains were analyzed statistically by the analysis of variance (Snedecor 1946).

Pigs fed the supplement containing the three B-vitamins gained faster than the pigs which received only the basal supplement; however, this increased rate of gain was not statistically significant. When 6 gm. terramycin and 6 mg. vitamin B_{12} per 100 lb. of supplement were added with the three B-vitamins (lot 3), gains were significantly increased over lots 1 and 2. Pigs that received antibiotic and vitamin B_{12} along with the three B-vitamins reached market weight (200 lb.) two weeks earlier than those receiving only the three B-vitamins and three weeks earlier than those receiving the basal ration alone.

Differences in consumption of corn and supplement by each lot are presented in table 2. Pigs fed the basal supplement plus the three

[3] Contained 3 gm. terramycin and 3 mg. vitamin B_{12} per lb. of supplement.

TABLE 1. COMPOSITION OF PROTEIN SUPPLEMENTS

(Trial 1)

Component	Lot 1 Basal	Lot 2 Basal + 3 B-Vitamins	Lot 3 Basal + 3 B-Vitamins + Antibiotic-B_{12} Supplement
	%	%	%
Tankage	90	90	88[1]
Alfalfa Meal	10	10	10
Antibiotic-B_{12} Supplement [2]	..	..	2
Total	100	100	100
Riboflavin (mg. per lb.)[3]		5.6	5.6
Calcium Pantothenate (mg. per lb.)		31.5	31.5
Niacin (mg. per lb.)		35.0	35.0

[1] 2% of tankage in supplement was replaced by the antibiotic-B_{12} supplement.
[2] Supplies 6 gm. terramycin and 6 mg. B_{12} per 100 lb. of protein supplement.
[3] Provides the recommended NRC allowances assuming the intake of corn and supplement to be 6:1.

B-vitamins along with terramycin and vitamin B_{12} consumed less supplement but utilized the total ration more efficiently than those on the basal treatment. In contrast, the pigs which received only the three B-vitamins in addition to the supplement consumed more supplement and were less efficient in their utilization of the total ration. These data suggest that vitamin B_{12} and terramycin are more effective in extending the protein supplement than are the three B-vitamins.

TABLE 2. EFFECT OF ADDING THREE B-VITAMINS, TERRAMYCIN, AND B_{12} TO A RATION FOR GROWING-FATTENING PIGS IN DRY LOT

(Trial 1)

Items Compared	Lot 1 Basal	Lot 2 Basal + 3 B-Vitamins	Lot 3 Basal + 3 B-Vitamins + Terramycin and B_{12}
Pigs per lot [1]	8[2]	9	10
Experimental period, wk.	17	16	14
Av. Initial wt., lb.	32.7	32.8	32.5
Av. Final wt., lb.	200.3	200.0	201.1
Av. daily gain, lb.[3]	1.30	1.46	1.72
Av. daily corn, lb.	3.74	4.55	4.86
Av. daily supplement, lb.	0.83	0.90	0.73
Feed per 100 lb. gain, lb.	351.4	373.7	325.3

[1] Equal numbers of barrows and gilts in each lot.
[2] Two pigs in lot 1 and one pig in lot 2 died after 8 weeks on experiment.
[3] LSMD, 5%, 0.21; 1%, 0.28.

Trial 2

Experimental Procedure

Thirty weanling crossbred pigs with an average initial weight of 36 lb. were segregated according to sex. The male and female pigs were then classified into outcome groups of 5 each and were alloted at random to 6 treatments in such manner that there were 2 males and 3 females in each treatment. The pigs were housed individually in

TABLE 3. COMPOSITION OF ANIMAL PROTEIN BASAL RATIONS
(Trial 2)

Component	Liveweight of pigs		
	Weaning to 100 lb.	100 to 150 lb.	150 to 200 lb.
	Percent of Ration		
Ground Yellow Corn	82.75	86.25	89.75
Tankage [1]	16.75	13.25	9.75
Iodized Salt	.50	.50	.50
	100.00	100.00	100.00
Vitamin A and D oil,[2] gm./100 lb.	60.0	60.0	75.0

[1] 65.5% protein (N × 6.25).
[2] 2250 I.U. Vitamin A and 400 I.U. Vitamin D per gram.

indoor pens with concrete floors. All pigs were hand fed for a period of 99 days which was the length of time required for pigs on the most rapidly growing treatment to reach an average weight of 200 lb.

Composition of the all-aminal protein basal rations is given in table 3. These rations were calculated to meet the recommendations of the National Research Council (1950).

Pigs assigned to treatment 1 received the basal ration. The pigs in each successive treatment received this ration plus vitamins and antibiotic in combinations described below, over the entire experimental period. Pigs in treatment 2 were given 0.8 mg. riboflavin, 4.5 mg. calcium pantothenate and 5.0 mg. niacin per lb. of feed. Pigs in treatment 3 received vitamin B_{12}[4] at the rate of 10 mcg. per lb. of feed; in treatment 4, 5.0 mg. of terramycin [5] were added per lb. of feed. The same levels of vitamin B_{12} and terramycin were added to the rations in treatment 5, and a combination of the three B-vitamins, vitamin B_{12}, and terramycin was fed to the pigs in treatment 6.

[4] Supplied by vitamin B_{12} supplement having a potency of 12.5 mg. vitamin B_{12} activity per lb.
[5] Supplement contained 5.0 gm. terramycin per lb. of supplement.

Results

The effects of supplementing a corn-tankage ration with vitamins and antibiotic are summarized in table 4; there was no apparent difference in the performance of males and females. The addition of the three B-vitamins to the basal ration markedly increased growth and improved appetite and feed efficiency. Similarly, the addition of vitamin B_{12} and/or terramycin improved growth, appetite, and feed efficiency. A combination of vitamin B_{12}, terramycin, and the three B-vitamins provided the greatest growth response observed in this trial. Responses of pigs that were fed these different dietary essentials in addition to the basal ration show that corn-tankage rations are not adequate for optimum development of pigs. These data suggest that the combination of certain B-vitamins may be a major limiting factor.

Differences in growth rates between lots were consistent throughout the experiment. Therefore, the addition of these different nutritional essentials is necessary in the later period as well as in the early growth period.

Occurrence of Pantothenic Acid Deficiency. After 58 days on trial (liveweight—110 lb.), symptoms of pantothenic acid deficiency were first noted in treatment 3 (basal plus vitamin B_{12}). According to the report by Yacowitz *et al.* (1951), vitamin B_{12} exerts a sparing action on the pantothenic acid requirement of chicks; however, the results of this trial, which agree with reports by Luecke *et al.* (1952), do not suggest that the same relationship exists with pigs. Luecke *et al.* (1952) reported that pantothenic acid deficiency could be produced on a plant protein ration that supplied 14 percent protein, and they suggested that higher protein levels spare the pantothenic acid requirement. However, in this experiment (trial 2) 18 percent protein failed to prevent deficiency symptoms when the basal ration supplied 3.5 mg. pantothenic acid per lb. Two pigs receiving terramycin supplementation began goose-stepping after 65 days (liveweight—100 lb.) on treatment (figure 1). Goose-stepping was also produced in treatment 3 where only vitamin B_{12} was added to the diet. No deficiency symptoms were noted when vitamin B_{12} and terramycin were administered together. Although goose-stepping appeared earlier in the vitamin B_{12} and terramycin-treated groups, the condition was observed in three pigs that received the basal diet. Apparently the growth stimulation produced by vitamin B_{12} and terramycin supplementation increased the requirement for pantothenic acid, and, therefore, induced deficiency symptoms earlier. This observation does not explain why deficiency symptoms

TABLE 4. SUPPLEMENTARY VALUE OF THREE B-VITAMINS, B_{12}, AND TERRAMYCIN IN A CORN-TANKAGE RATION FOR GROWING PIGS

(Trial 2)

	Treatment					
Comparisons	1 Basal	2 Basal + 3 B-Vitamins	3 Basal + B_{12}	4 Basal + Terramycin	5 Basal + B_{12} + Terramycin	6 Basal + B_{12} + Terramycin + 3 B-Vitamins
Number of Animals [1]	4 [2]	5	5	5	5	5
Av. Initial Weight, lb.	36.0	36.0	36.2	36.0	35.8	35.8
Av. Final Weight, lb.	128.8	176.4	145.8	153.8	171.5	200.0
Av. Daily Gain, lb.[3]	.84	1.42	1.11	1.19	1.37	1.66
Av. Daily Feed, lb.	3.97	4.67	4.53	4.58	4.66	5.59
Feed per 100 lb. gain, lb.	472.1	329.3	408.9	384.9	340.3	337.0

[1] Three gilts and two barrows per treatment.
[2] One pig developed a respiratory disorder and died after 74 days on treatment.
[3] LSMD, 5%, 0.29; 1%, 0.39.

were not produced when vitamin B_{12} and terramycin were administered together. Supplementing the diet with calcium pantothenate (4.5 mg. per lb. of feed) prevented the condition in treatments 2 and 6.

After termination of the experiment, all goose-stepping pigs were placed on the basal ration which was then supplemented with 1.6, 9.0,

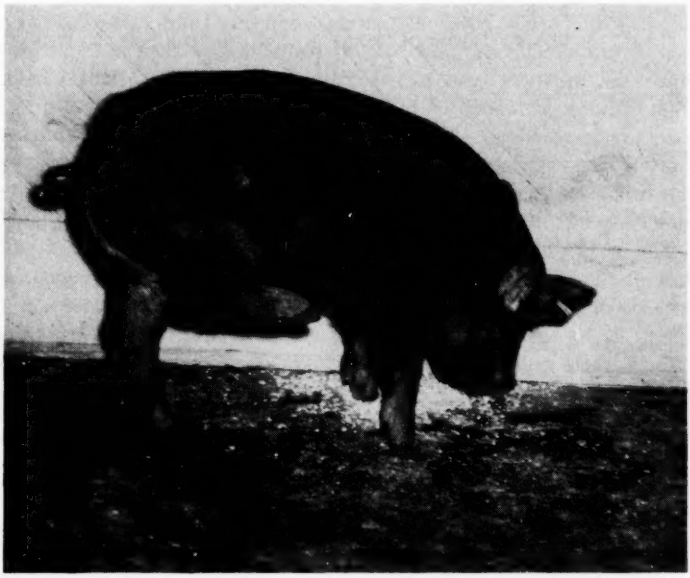

Figure 1. Severe "goose-stepping" observed after 70 days on terramycin treatment (Lot 4, Trial 2).

and 10.0 mg. of riboflavin, calcium pantothenate, and niacin, respectively. This vitamin therapy completely cured deficiency symptoms of short duration (basal treatment). Even the most severely deficient pigs responded in some degree to such treatment; but after a period of 70 days, the condition continued to persist. Evidently the prolonged deficiency state had caused irreparable damage.

Trial 3

Experimental Procedure.

The purpose of this experiment was to determine the supplemental effect of lysine, methionine, and tryptophan upon two basal rations,

one containing tankage and the other containing meat and bone scraps.

The basal rations were formulated to provide similar protein levels which remained the same throughout the experimental period and were purposely restricted to 14.5 percent total protein to provide a more critical evaluation of the supplementary effect of the amino acids. The basal rations given in table 5 were calculated to satisfy the rec-

TABLE 5. COMPOSITION OF MEAT BY-PRODUCT BASAL RATIONS

(Trial 3)

Component	Liveweight of Pigs Weaning to 125 lb.	
	Tankage Basal	Meat Scrap Basal
	Percent of Ration	
Ground Yellow Corn	85.50	83.75
Digester Tankage [1]	14.00	
Meat and Bone Scrap [2]		15.75
Salt	.50	.50
	100.00	100.00
Supplements per 100 lb.:		
Vitamin A and D oil,[3] gm.	60.00	60.00
Riboflavin, mg.	80.00	80.00
Calcium Pantothenate, mg	450.00	450.00
Niacin, mg.	500.00	500.00
Vitamin B_{12} Supplement,[4] gm.	90.80	90.80
Terramycin Supplement,[5] gm.	68.10	68.10

[1] 62.2% protein (N × 6.25).
[2] 55.6% protein (N × 6.25).
[3] 2250 I.U. vitamin A and 400 I.U. vitamin D per gram.
[4] Provides 5 mg. vitamin B_{12} per lb.
[5] Provides 5 gm. terramycin per lb.

ommended allowances of the National Research Council for the other essential nutritional elements. All rations supplied 10 mcg. vitamin B_{12} and 7.5 mg. terramycin per lb. of feed.

Six healthy weanling Duroc pigs were placed on each of seven experimental treatments. Two pigs, a gilt and a barrow, were placed in a 4 by 7 foot concrete floored pen. Three such replications constituted a single experimental treatment. The two pigs in each pen were self-fed the total ration by means of an individual type self-feeder. Pigs in all replications were fed until they reached an average weight of 125 lb.

Pigs on treatments 1 and 2 received the basal rations given in table 5. To the tankage basal was added 0.6 percent DL-lysine (treatment 3); 0.2 percent DL-methionine (treatment 4); and 0.1 percent DL-tryptophan (treatment 5). Pigs in treatment 6 received the same

basal to which was added the above quantities of each of the amino acids. In treatment 7 this same amino acid combination was added to the meat scrap basal.

Results

Pigs fed the tankage basal grew faster than those which were fed a similar ration containing meat scraps. However, when both rations were supplemented with all three amino acids, this difference was not noted (table 6). Apparently the deficiencies of meat scraps are more severe than those in tankage. It is difficult to determine whether this is a single or multiple deficiency because the meat scrap basal was not supplemented with single amino acids; but supplementing meat scraps with lysine, methionine, and tryptophan increased growth significantly. Supplementation of the tankage basal with lysine failed to stimulate rate of gain or feed efficiency. Methionine depressed gain and appetite as compared to the controls, but this difference was not significant.

Tryptophan improved the tankage basal notably with respect to gain, appetite, and feed efficiency, although a combination of all three amino acids afforded no more stimulation than tryptophan alone. These data suggest that tryptophan is the primary limiting amino acid in a corn-tankage ration.

Discussion

The results obtained in this series of feeding trials using corn-meat by-product rations suggest that animal proteins must be supplemented with vitamin B_{12} if optimum growth rate and feed efficiency are to be realized. In the first two trials of this series the growth rate was increased by the addition of B-complex vitamins which shows that B-vitamins are needed with animal as well as plant proteins (Briggs and Beeson, 1951, 1952, and Luecke *et al.*, 1951). In the first experiment the greatest growth response was obtained by supplementation with terramycin and B_{12}. Even though response to the addition of the B-vitamins alone was not significant (trial 1), the presence of these vitamins in combination with terramycin and B_{12} undoubtedly contributed to the marked effect of this treatment upon growth response and feed efficiency. Briggs and Beeson (1951) observed that the addition of B-vitamins to a plant and animal protein basal containing antibiotic and vitamin B_{12} gave additional growth stimulation. In the second trial of this series a similar observation was made when an animal protein basal was used.

TABLE 6. PIG RESPONSE TO AMINO ACID SUPPLEMENTATION OF CORN-TANKAGE AND CORN-MEAT SCRAP RATIONS FORTIFIED WITH B-VITAMINS AND ANTIBIOTIC

(Trial 3)

Comparisons	1 Tankage Basal	2 Meat Scrap Basal	3 Tankage Basal + Lysine	4 Tankage Basal + Methionine	5 Tankage Basal + Tryptophan	6 Tankage Basal + Lysine + Methionine + Tryptophan	7 Meat Scrap Basal + Lysine + Methionine + Tryptophan
Number of Animals [1]	6	5 [2]	6	6	6	6	6
Av. Initial Weight, lb.	29.8	30.6	29.8	29.7	29.7	29.7	29.8
Av. Final Weight, lb.	127.2	125.2	125.5	127.2	125.3	126.3	125.9
Av. Daily Gain, lb.	1.39	1.13	1.35	1.27	1.54	1.51	1.50 [3]
Av. Daily Feed, lb.	4.44	3.88	4.27	4.08	4.81	4.86	4.60
Feed per 100 lb. gain, lb.	319.5	344.6	317.1	322.4	311.7	321.9	306.7 [4]
Days on experiment	70	84	71	77	62	64	64

[1] Three barrows and 3 gilts per treatment.
[2] One pig was removed because of extreme unthriftiness.
[3] LSMD, 5%, 0.31; 1%, 0.42.
[4] LSMD, 5%, 28.4; 1%, 39.8.

Brinegar *et al.* (1950), Shelton *et al.* (1951a, 1951b, 1951c), and Curtin *et al.* (1952) have reported the lysine, methionine, and tryptophan requirements for normal growth of pigs. Corn is recognized as being a limiting source of lysine and tryptophan while meat products provide greater quantities of these amino acids (Block and Bolling, 1951). Results of the third investigation in this series imply that only tryptophan may be of a limiting nature in corn-tankage rations. Tankage used in this trial contained more tryptophan (0.43 percent) as determined microbiologically by the method outlined by Steele *et al.* (1949), than did meat and bone scraps (0.30 percent). Therefore, it is logical to assume that tryptophan also is the most notably limiting amino acid in meat scraps. Bloss *et al.* (1953) reported the supplementary value of tryptophan in corn meat scrap rations. The difference in response of pigs fed tankage compared to those fed meat scraps may be partially accounted for by such existing differences in the quantity of essential amino acids in the basal rations. Growth rate was equalized by adding lysine, methionine, and tryptophan (trial 3, table 6) to either the meat scrap (treatment 7) or tankage (treatment 6) diets.

Calculated values for tryptophan in the basal rations were 0.097 and 0.084 percent for the tankage basal and meat scrap basal respectively. Shelton *et al.* (1951c) reported the requirement for pigs to be 0.2 percent DL-tryptophan. Since the D-isomer of tryptophan is partially utilized (Thompson *et al.*, 1952), the requirement then must exceed 0.1 percent L-tryptophan. Therefore the amount of tryptophan supplied by the tankage (0.097 percent) and meat scrap (0.084 percent) basals was below the requirement. When DL-tryptophan was added, the needs for this amino acid were supplied and growth rate was increased.

The tankage basal and meat scrap basal provided 0.63 and 0.72 percent L-lysine respectively. Although the tankage basal was deficient in lysine according to the requirements (1.0 percent L-lysine) reported by Shelton *et al.* (1951a) and (1.2 percent L-lysine) Brinegar *et al.* (1950), there was no stimulation when additional lysine was provided. It is not likely that the limited tryptophan content of the tankage basal prevented a response to additional lysine. This suggestion is supported by the fact that lysine gave no response when an adequate level of tryptophan was supplied (treatment 6).

The approximate content of methionine plus cystine was 0.35 percent in the tankage basal and 0.44 percent in the meat scrap basal. Such values were below the required levels. (0.6 percent DL-methionine)

reported by Shelton *et al.* (1951b) and (0.7 percent DL-methionine) reported by Curtin *et al.* (1952). Additional methionine, however, produced a growth inhibition although it was not significant. A similar observation when excess methionine was fed to rats was reported by Steffee *et al.* (1950). The observed growth depression produced by methionine supplementation may have been conditioned by limited tryptophan in the basal ration. When methionine was added to a ration properly supplemented with tryptophan (treatment 6), this growth inhibition was not noted.

Microbiological assay revealed that the meat scraps used in this trial provided more lysine and methionine than tankage. Since tankage contains sufficient quantities of these amino acids, it may be assumed that meat scraps are likewise adequate.

Summary

Three experiments were conducted to determine the value of supplementing corn-meat by-product rations with vitamins, antibiotic, and amino acids. Results of the first trial presented evidence that vitamin B_{12} and terramycin stimulate growth.

In the second trial, pig response clearly indicated the supplementary values of combined riboflavin, calcium pantothenate, and niacin; of vitamin B_{12}; and of terramycin. Administration of the three B-vitamins afforded greater response than either vitamin B_{12} or terramycin. These factors in combination with the three B-vitamins stimulated development still further.

Pantothenic acid deficiency symptoms occurred in pigs receiving vitamin B_{12}, those receiving terramycin supplementation, and in individuals receiving the basal ration. Administration of increased levels of pantothenic acid effected a cure in the latter group, but failed to completely allay the condition in the vitamin B_{12} or terramycin-treated groups.

Results of the third trial indicate that corn-tankage rations are adequate in lysine and methionine, but limiting in tryptophan. Rations containing meat scraps as the only protein supplement are inadequate to sustain optimum growth. Tryptophan is probably the most limiting amino acid in a corn meat and bone scrap ration.

Literature Cited

Anderson, G. C. and A. G. Hogan. 1949. A deficiency of Vitamin B_{12} in a practical ration for weanling pigs. JOURNAL OF ANIMAL SCIENCE 8:614.

Blight, J. C. and W. C. Powick. 1951. The value of methionine, lysine and vitamin B_{12} as supplements to a corn-soybean meal diet for pigs and rats. JOURNAL OF ANIMAL SCIENCE 10:1039.

Block, R. J. and D. Bolling. 1951. The amino acid composition of proteins and foods. 2nd Edition. Charles C. Thomas, Publisher, Springfield, Ill.

Bloss, R. E., R. W. Luecke, J. A. Hoefer, F. Thorp, Jr., and W. N. McMillen. 1953. Supplementation of a corn-meat and bone scrap ration for weanling pigs. JOURNAL OF ANIMAL SCIENCE 12:102

Briggs, J. E. and W. M. Beeson. 1951. The supplementary value of riboflavin, calcium pantothenate and niacin in a practical mixed animal and plant protein ration containing vitamin B_{12} and aureomycin for weanling pigs in dry lot. JOURNAL OF ANIMAL SCIENCE 10:813.

Briggs, J. E. and W. M. Beeson. 1952. The effect of vitamin B_{12}, aureomycin, streptomycin or dried whey factor supplements on the growth and fattening of weanling pigs. JOURNAL OF ANIMAL SCIENCE 11:103.

Brinegar, M. J., H. H. Williams, J. K. Loosli, and L. A. Maynard. 1950. Lysine requirement for growing swine. Fed. Proc. 9:353.

Burnside, J. E., T. J. Cunha, A. M. Pearson, R. S. Glasscock, and A. L. Shealy. 1949. Effect of APF supplements on pigs fed different protein supplements. Arch. Biochem. 23:328.

Catron, D. V. and C. C. Culbertson. 1949. Faster gains with APF. Iowa Farm Science 3:3.

Curtin, L. V., J. K. Loosli, J. Abraham, H. H. Williams, and L. A. Maynard. 1952. The methionine requirement for the growth of swine. JOURNAL OF ANIMAL SCIENCE 11:759.

Eggert, R. G., M. J. Brinegar, and C. R. Anderson. 1951. The effect of supplementing normal and high protein corn with lysine and tryptophan for swine. JOURNAL OF ANIMAL SCIENCE 10:1047.

Hart, E. B. and H. Steenbock. 1919. Maintenance and production value of some protein mixtures. Jour. Biol. Chem. 38:267.

Lepley, K. C., D. V. Catron, and C. C. Culbertson. 1950. Dried whole aureomycin mash and meat and bone scraps for growing-fattening swine. JOURNAL OF ANIMAL SCIENCE 9:608.

Luecke, R. W., F. Thorp, Jr., H. W. Newland, and W. N. McMillen. 1951. The growth promoting effects of various antibiotics on pigs. JOURNAL OF ANIMAL SCIENCE 10:538.

Luecke, R. W., J. A. Hoefer, and F. Thorp, Jr. 1952. The relationship of protein to pantothenic acid and vitamin B_{12} in the growing pig. JOURNAL OF ANIMAL SCIENCE 11:238.

Noland, P. R., J. P. William, and F. B. Morrison. 1951. Vitamin and mineral supplements for growing and fattening pigs in drylot. JOURNAL OF ANIMAL SCIENCE 10:875.

National Research Council. 1950. Recommended nutrient allowances for swine. No. 2:3.

Shelton, D. C., W. M. Beeson, and E. T. Mertz. 1951a. Quantitative L-lysine requirement of the weanling pig. Arch. Biochem. 30:1.

Shelton, D. C., W. M. Beeson, and E. T. Mertz. 1951b. The effect of methionine and cystine on the growth of weanling pig. JOURNAL OF ANIMAL SCIENCE 10:57.

Shelton, D. C., W. M. Beeson, and E. T. Mertz. 1951c. Quantitative DL-tryptophan requirement of the weanling pig. JOURNAL OF ANIMAL SCIENCE 10:73.

Snedecor, G. W. 1946. Statistical methods. 4th Edition. Iowa State College Press, Ames, Iowa.

Steele, B. F., H. E. Sauberlich, M. S. Reynolds and C. A. Bauman. 1949. Media for leuconostoc mesenteroides P-60 and leuconostoc citrovorum 8081. Jour Biol. Chem. 177:533.

Steffee, C. H., R. W. Wissler, E. M. Humphreys, E. P. Benditt, R. W. Woolridge, and P. R. Cannon. 1950. Studies in amino acid utilization. V. The determination of minimum daily essential amino acid requirements in protein-depleted adult male albino rats. Jour. Nutr. 40:483.

Thompson, C. M., E. Reber, C. K. Whitehair, and R. MacVicar. 1952. Utilization of D-tryptophan by swine. JOURNAL OF ANIMAL SCIENCE 11:712.

Vestal, C. M. 1946. Mixed supplements for hogs. Ind. Ag. Exp. Sta. Bul. No. 508.

Yacowitz, H., L. C. Norris, and G. F. Heuser. 1951. Evidence for an interrelationship between vitamin B_{12} and pantothenic acid. Jour. Biol. Chem. 192:141.

EFFECT OF THREE LEVELS OF A NEW ANTIBIOTIC, TETRACYCLINE, IN A SWINE RATION [1]

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THE growth response produced by certain antibiotics, notably chlortetracycline (Aureomycin) and oxytetracycline (Terramycin), in the rations of growing-fattening swine is well established. A new antibiotic, tetracycline, has been produced by the reduction of chlortetracycline (Boothe *et al.*, 1953) and also by the reduction of oxytetracycline (Conover *et al.*, 1953). A feeding trial was conducted, therefore to compare several levels of tetracycline.

Procedure

Thirty Duroc weanling pigs, 15 gilts and 15 barrows, were divided on the basis of sex and weight into five lots of six pigs each. Group weights were recorded at 2-week intervals. Individual weights were recorded at the beginning of the trial, at the time the pigs of each group reached an average weight of approximately 100 lb., and at the end of the trial. Feed intake was recorded at the 100-lb. average point and at the termination of the trial.

The basal ration consisted of 58.4 percent corn, 22 percent barley, 7.5 percent soybean oil meal, 5 percent meat scraps, 5 percent alfalfa meal, 2 percent mineral supplement, and 0.1 percent vitamin B-complex.[3] The mineral supplement contained 0.75 lb. salt, 0.50 lb. ground limestone, 0.50 lb. steamed bone meal, and 0.25 lb. "Mico" trace mineral mixture.

Lot 1, the negative control, was fed the basal ration alone; Lots 2, 3, and 4, received the basal ration supplemented with 10.85, 15.03, and 19.21 gm. of tetracycline per ton, respectively. The positive control, lot 5, received the basal ration supplemented with 18 gm. chlortetracycline and 18 mg. B_{12} per ton.

The source of the tetracycline was a soya flour supplement containing 0.835 gm. anhydrous tetracycline base per lb. and the source of the

[1] Journal Series paper of the New Jersey Agricultural Experiment Station, Rutgers University, The State University of New Jersey, Department of Animal Husbandry, New Brunswick.

[2] Part of the data presented herein was taken from a thesis presented by D. J. Horvath to the graduate faculty of Rutgers University in partial fulfillment of the requirement for the degree of Master of Science.

[3] The vitamin B supplement (No. 58-A) was furnished by Merck and Company, Rahway, N. J., through the courtesy of Dr. H. Draper.

chlortetracycline was Aurofac, supplied by Lederle Laboratories, Pearl River, New York, through the courtesy of Dr. Ralph J. Elliott.

Results and Discussion

The average daily gains and feed efficiency of the pigs in the different lots are shown in table 1; a comparison of these figures as percent of the negative control appears in table 2.

Initial Weight to 100 Pounds

The effect of the antibiotics was most marked in the first part of the trial. All lots of pigs except one (lot 2) made slightly greater gains than the negative control lot. The positive control lot and the lot fed the 15.03 gm. level of tetracycline made the most rapid gains, each exceeding the negative control lot by 3.3 percent. The greatest differences, however, occurred in feed conversion; the positive and negative control lots being almost identical, whereas the tetracycline lots averaged 20.2 percent less feed per lb. of gain. The 15.03 gm. level of tetracycline was most efficient, since pigs in that lot used only 68.6 percent as much feed per lb. of gain as the negative controls.

100 Pounds to Market Weight

In the second part of the trial, the negative control lot gained most rapidly, and the positive control, paradoxically, gained least rapidly. Of the three lots fed tetracycline, the one fed the intermediate level again made greatest daily gains. Although the relative differences in feed efficiency in this period were less than in the first period, lots fed the two lower levels of tetracycline, particularly the 15.03 gm. level, again led in feed efficiency. The lot fed the latter level had an advantage of 5.4 percent compared to the negative control, and 9.3 percent compared to the positive control.

Initial Weight to Market Weight

Average gain of the lot fed the 15.03 gm. level of tetracycline exceeded that of the other lots fed antibiotics but was 1.6 percent less than that of the negative control lot. Feed efficiency was increased at all three levels of tetracycline supplementation. The 15.03 gm. level was most efficient, pigs in that lot requiring 10.6 percent less feed than those in the negative control lot. The positive control lot of pigs fed

chlortetracycline required 3.7 percent more feed per pound of gain than the negative control and 16.1 percent more than those fed the 15.03 gm. level of tetracycline.

TABLE 1. EFFECT OF TETRACYCLINE ON RATE OF GAIN AND FEED EFFICIENCY OF PIGS AT DIFFERENT WEIGHTS

(Six pigs per lot)

Lot	Antibiotic Supplement[1]	Average Initial Wt.	Standard Error	Average Daily Gain[2]	Feed per lb. Gain	Days	Average Final Weight	Standard Error
		Initial weight to 100 pounds						
		lb.		lb.	lb.		lb.	
1	None	46.50	3.6	1.82	2.99	33	108.33	4.8
2	10.85 gm. tetracycline	45.66	2.2	1.79	2.79	31	101.00	4.0
3	15.03 gm. tetracycline	46.33	2.7	1.88	2.05	28	99.00	5.4
4	19.21 gm. tetracycline	45.83	1.5	1.84	2.32	31	102.83	2.6
5	18 gm. chlortetracycline + 18 mg. B_{12}	45.50	3.9	1.88	3.02	31	103.66	6.4
		100 lb. to market weight (200 lb.)						
1	None	108.33	4.8	1.94	4.24	51	207.16	5.4
2	10.85 gm. tetracycline	101.00	4.0	1.85	4.09	53	199.16	4.5
3	15.03 gm. tetracycline	99.00	5.4	1.88	4.01	56	204.50	7.7
4	19.21 gm. tetracycline	102.83	2.6	1.84	4.29	53	206.66	5.6
5	18 gm. chlortetracycline + 18 mg. B_{12}	103.66	6.4	1.84	4.42	53	201.66	6.5
		Initial weight to market weight						
1	None	46.50	3.6	1.91	3.76	84	207.16	5.4
2	10.85 gm. tetracycline	45.66	2.2	1.83	3.62	84	199.16	4.5
3	15.03 gm. tetracycline	46.33	2.7	1.88	3.36	84	204.50	7.7
4	19.21 gm. tetracycline	45.83	1.5	1.84	3.57	84	206.66	5.6
5	18 gm. chlortetracycline + 18 mg. B_{12}	45.50	3.9	1.86	3.90	84	201.66	6.5

[1] Values are per ton of feed.

[2] Analysis of variance (Paterson, 1939) of the individual average daily gains for each period indicated no significant differences between the treatments at the 5% level.

In general, the supplements did not elicit the growth response usually reported for antibiotics. The relatively rapid gains and the absence of scouring in all lots, however, were indications of an adequate nutritional and health status. This condition would preclude to some degree the possibility of antibiotic growth-stimulation. Of the levels of tetra-

cycline tried, the 15-gm. level appeared optimum in terms of gains and feed efficiency.

In this trial and another recently completed one, chlortetracycline failed to exert effects observed in earlier experiments. A decline in relative response has developed after two years of almost continuous

TABLE 2. EFFECT OF TETRACYCLINE ON RATE OF GAIN AND FEED EFFICIENCY OF SWINE EXPRESSED AS PERCENT OF THE NEGATIVE CONTROL

		Initial Wt.-100 lb.		100 lb.-Market Weight		Initial Wt.-Market Weight	
Lot	Antibiotic Supplement	Average Daily Gain	Feed per lb. Gain	Average Daily Gain	Feed per lb. Gain	Average Daily Gain	Feed per lb. Gain
1	None	100.0	100.0	100.0	100.0	100.0	100.0
2	10.85 gm. tetracycline	98.4	93.3	95.4	96.5	95.8	96.3
3	15.03 gm. tetracycline	103.3	68.6	96.9	94.6	98.4	89.4
4	19.21 gm. tetracycline	101.1	77.6	94.9	101.2	96.3	95.0
5	18 gm. chlor-tetracycline + 18 mg. B_{12}	103.3	101.0	94.9	104.2	97.4	103.7

chlortetracycline feeding, but may not occur where it has not been used so extensively. The decline in relative response was associated with a steady increase in gains made in successive years by the lots fed the basal ration. In the present trial an unexplained depression in growth during the last two weeks of the second period reduced the advantage in rate of gain from chlortetracycline.

Summary

An experiment was conducted with 30 Duroc pigs to compare three levels of tetracycline in a swine ration to a recommended level of chlortetracycline and a negative control. The levels of tetracycline were 10.85, 15.03, and 19.21 gm. per ton.

1. Tetracycline at all levels used improved feed efficiency but did not improve rate of gains as compared to chlortetracycline and to the negative control. The increase in feed efficiency was greatest in the initial part of the trial.

2. The optimal level of tetracycline appeared to be 15 gm. per ton of feed.

3. Chlortetracycline failed to produce the responses observed in previous trials. This was associated with improved performance of the pigs on basal rations.

Literature Cited

Boothe, J. H., J. Morton, J. P. Petisi, R. G. Wilkinson and J. H. Williams. 1953. Tetracyline. Jour. Am. Chem. Soc. 75:4621.

Conover, L. H., W. T. Moreland, A. R. English, C. R. Stephens and F. J. Pilgrim. 1953. Terramycin XI, Tetracyclin. Jour. Am. Chem. Soc. 75:4622.

Paterson, D. D. 1939. Statistical technique in agricultural research. McGraw-Hill, Inc., New York.

THE EFFECT OF ANTIBIOTICS ON THE INTESTINAL AND CECAL MICROFLORA OF BABY PIGS FED A "SYNTHETIC MILK"

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IN seeking an explanation for the stimulation of growth in animals fed antibiotic substances, numerous investigators have attempted to find significant changes in the intestinal microflora. The results of these investigations have, for the most part, been negative. Braude, Kon and Porter, in their excellent review article (1953) have suggested a number of explanations for these negative results.

Work done previously at this laboratory by Wahlstrom, *et al.* (1950, 1951, 1952) has demonstrated that the effect of aureomycin, chloromycetin, penicillin and sulfathalidine on numbers of *Escherichia coli,* lactobacilli and yeasts is not correlated consistently with the growth response.

All of these studies were conducted on fecal samples and thus gave no information on the flora in other sections of the gut. We chose to investigate the effect of two antibiotics, terramycin, an antibacterial agent, and rimocidin, an antibiotic effective against yeast and molds, on the microflora higher in the intestine and cecum of the baby pig fed a "synthetic milk." Two levels of B vitamins were also tested in these experiments.

The effect of these treatments on growth has been reported and discussed in an earlier paper by Schendel and Johnson (1953) in which terramycin was shown to exert a significant effect.

Experimental

Sixteen baby pigs were removed from their dams at 48 to 96 hours of age and assigned at random to 8 treatment groups. The design used was a 2^3 factorial design, the three factors being level of B vitamin supplementation and the presence or absence of terramycin

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and of rimocidin. The animals were maintained throughout the experimental period in raised, wire-bottom cages and fed *ad libitum*. Individual feed consumption and weight records were kept throughout the experiment.

The composition of the 30 percent alpha-protein "synthetic milk" has been described by Schendel and Johnson (1953). Eight of the animals received the water soluble vitamins at a level 5 times the amount used previously in this laboratory and with which we have been able to demonstrate growth stimulation using several antibiotics (Nesheim, Krider and Johnson, 1950) (Wahlstrom, Terrill and Johnson, 1950) (Wahlstrom and Johnson, 1951). The method of feeding and care of baby pigs in this laboratory has also been reported (Johnson, James and Krider, 1948).

The antibiotics were given at a level of 50 mg per kg dry matter of diet. These, as well as the two levels of water soluble vitamins, were added to the milk at the time of feeding.

When the animals reached 5 weeks of age they were sacrificed and bacterial plate counts were made of the contents of four sections of the small intestine, and of the cecum.

The animals were suspended by a hind limb and sacrificed by sectioning the jugular vein. As soon as the pig became quiet, having been restricted during this period of struggling so as to reduce the movement of gut contents in the tract, the section of the gut from the cardiac sphincter valve through the rectum was carefully excised. Then, with the intestines still intact, the stomach and cecum were isolated from the remainder of the gut by ligature and numerous "blind ligatures" were tied throughout all portions of the gut to confine the contents in their respective areas.

After the continuity of the mesentery had been destroyed and the intestine straightened, 4 of the previously isolated sections were selected to be representative of the duodenal, jejunal and ileal sections of the small intestine. The contents of these sections and of the cecum were then thoroughly evacuated into sterile graduated cylinders. The contents were weighed, made up to an appropriate volume, homogenized in a micro Waring blendor or with a mechanical stirrer, and plated out in triplicate on 4 types of media: beef liver infusion (two sets, one incubated aerobically, the other anaerobically), malt agar, Rogosa's medium and desoxycholate agar. Colonies were counted after incubation at 37° C for 48–72 hours.

Results and Discussion

The statistical analysis of the growth data was carried out by the method of fitting constants and has been discussed in an earlier paper (Schendel and Johnson, 1953). There was no stastically significant interaction of the three treatments, and none of the seven variables exerted a significant effect on the final weight with the exception of terramycin and feed consumption (respective P values of 0.038 and less than 0.01). The effect of terramycin on final weight after equalization of feed consumption (P less than 0.05) suggests this antibiotic exerts an effect of from 0.24 to 6.06 kg. over a five week period in addition to any effect of increasing feed consumption. The affect of rimocidin, two levels of B vitamins, the initial variation in weight and age as well as the source of animals on final weight was not found to be significant.

In attempting to choose small intestinal sections which would represent all areas of the organ and contain sufficient food material, we realized the following ranges: Section A=0.91 to 4.2 percent (with one exception of 15 percent); Section B=20.3 to 36.9 percent; Section C=48.9 to 59.9 percent; Section D=70.1 to 87.6 percent of the total intestinal length.

The means of the bacterial plate counts appear in table 1 and the individual counts in table 2.

The results of the microbiological study indicate that the antibiotic terramycin may have a significant effect on the numbers of microbes in some sections of the intestinal tract. The effect is to increase about tenfold the numbers of micro-organisms found in the section between the duodenum and the cecum. The interpretation of these data, which are shown in table 1, is made difficult by the large variation within treatments. However, in Sections B, C and D of the gut there is only one instance in which the counts in the terramycin treated pigs were lower than in a similar specimen from the basal pigs.

Earlier work with the rat revealed that these were also the sections of the gut at which the antibiotic concentration was greatest (table 3). The rats were fed a pantothenic acid-deficient diet supplemented with aureomycin at levels of 50, 200 and 3000 μg./gm. diet. The assays were carried out using the pad plate method (Grady and Williams, 1952).

We suggest that the duodenal sections should be considered separately since the flora is probably affected more by the nature of the gastric contents flowing into it than by the antibiotic. Previous work

by Wahlstrom *et al.* (1950, 1951, 1953), indicated no correlation between counts of the cecal flora and antibiotic growth response. Our data appear to confirm these findings.

No intensive investigations of the nature of the microflora were carried out. The media used in this study were chosen, however, in the

TABLE 1. MEAN SMALL INTESTINAL AND CECAL MICROFLORA OF 15 BABY PIGS GROWN ON 4 MEDIA [1, 2]

Medium	Section				
	A	B	C	D	E
Basal Animals [3]					
Liver Infusion (aerobic)	59	9.8	98 [4]	1060	7090
Liver Infusion (anaerobic)	30	6.1	68	1240	7730
Rogosa	8	3.1	15	27	290
Desoxycholate	49	1.1	14 [4]	437	1500
Malt Agar	3.7 [4]	2	14 [4]	68	1615
Terramycin Treated Animals					
Liver Infusion (aerobic)	53	500	800	2500	4700 [4]
Liver Infusion (anaerobic)	130	243	856	2520	7079
Rogosa	19	15	24	63 [4]	260
Desoxycholate	4	33	130	1100	1200
Malt Agar	6.5	28	3.2	445	1100

[1] All counts expressed in units of 10^5 organisms per ml. gut contents.

[2] Each figure represents the mean of 7 animals in the basal group and 8 in the treated group except those marked [4] which represent 6 and 7 animals respectively due to improper plate concentration.

[3] Included in this group are those piglets which received rimocidin since this antibiotic was shown to have no significant effect on growth.

[4] Six animals if basal; 7 if treated.

hope that any marked differences in the following types of microbes would be made apparent:

Liver infusion (anaerobic) — Total anaerobes
Liver infusion (aerobic) — Total aerobes
Rogosa medium — Lactic acid bacteria
Desoxycholate medium — Coliform bacteria
Malt agar — Yeasts

From the observations made it was clear that none of the various types of bacteria found in the gut of basal animals was conspicuously absent from the contents of the gut of animals receiving terramycin. Rather there seemed to be an overall increase in numbers of bacteria with the antibiotic. Thus the antibiotic might have exerted its effect by bringing about an increase in numbers of microorganisms capable of synthesizing nutrients present in limiting concentrations. In the face

TABLE 2. SMALL INTESTINAL AND CECAL MICROFLORA OF INDIVIDUAL BABY PIGS GROWN ON FIVE MEDIA[1]

	Basal animals section: small intestine						Terramycin-treated animals section: small intestine				
Pig No.	A	B	C	D	Cecum	Pig No.	A	B	C	D	Cecum
Liver (aerobic) medium											
1	250	0.2	0.3	860		8	32	84	1	590	4200
2			410	570	9600	9	200	120	110	2300	7600
3	4.5	17	7.9	620	24000	10	1.5	0.1	1.1	150	1230
4		12	66	420	820	11	31	38	24	102	3900
5	4.5	16	13	190	2000	12	0.4	14	21	590	2740
6	15	1.7		71	3400	13	96	370	580	3900	2200
7	22	12	91	4700	2700	14	7.2	74	1100	9600	
						15	56	3300	4600	2700	11000
Liver (anaerobic) medium											
1	120	0.03	0.09	760	820	8	12	130	2000	558	6200
2	10	0.001	310	450	13200	9	770	103	142	1111	13100
3	4.2	17	7.2	470	20000	10	2.6	0.1	1.4	108	1260
4	12	8	74	390	820	11	29	44	46	48	2400
5	6.4	5.7	12	290	1880	12	1.1	11	1.9	560	1960
6	13	2.3	0.3	208	4800	13	43	220	580	7700	
7	32	9.5	75	5120	2600	14	4.8	67	820	7000	150
						15	178	1365	3240	3080	9700

TABLE 2.—*Continued*

Rogosa medium											
1	25	0.03		11	11	8	4.5	32	0.001	190	610
2	2.1	0.01	38	28	400	9	91	40	118	118	680
3	5.8	12	9.9	72	910	10	0.55	0.03	0.53	7.8	8
4	5.0	3.8	12	20	24	11	14	10	8.8	25	290
5	0.65	1.1	7.4	37	440	12		4.9	20	45	135
6	5.3	2.7		17	180	13	10	3.8	24	28	144
7	12	2.3	7.3	5.6	68	14	2.4	17	17	28	78
						15	5.8	10	7.5		133
Desoxycholate medium											
1	5.6	0.001	0.1	84	315	8	9.9	1.0	1	60	1700
2	2.4	0.001	36	39	2200	9	0.1	0.1	0.1	12.6	650
3	0.3	1.5	2.2	33	5600	10	0.8	0.01	0.9	29	1020
4	0.7	1.0	18.9	123	320	11	2	0.01	2	7.8	780
5	1.4	0.6	1.1	5.7	260	12	0.5	7	12	214	650
6	0.4	0.03		4	804	13	12	200	320	2700	920
7	7.8	2.6	27	2770	1000	14	4.3	19	540	5700	3800
						15	1.8	33	170	348	107
Malt agar medium											
1	11	0.07	0.2	123	87	8	12.6	2.8	0.4	10	1200
2	3.2	0.001	56	81	2000	9	1.7	3.6	2.2	14	770
3	1.8	6.8	4.2	56	5000	10	0.8	0.1	0.8	9	270
4		1.1	17.4	131	335	11	0.3	3.5	1.3	7	940
5	1.7	8.1	5.9	60	3100	12	0.4	8.5	10.4	14	820
6	0.7	0.08	0.009	18	776	13	24	180	10	3000	750
7	3.6	0.04		6.4	7.9	14	6.3	22	0.1	500	4100
						15	5.8	0.5	0.6	3.9	12

[1] All counts expressed in units of 10^5 organisms per ml. gut contents.

of this increase in total microbial population it seems less likely that the growth stimulus resulted from an inhibition of microorganisms producing toxic substances and/or those which may compete with the host for essential nutrients.

In later experiments using weanling rats (1954), we have observed reduced specific activities of carcass radiothiamin when it was fed

TABLE 3. CONCENTRATION OF AUREOMYCIN IN VARIOUS LEVELS OF THE SMALL INTESTINE

Section of Small Intestine	Level of Aureomycin Fed/gm. Diet			
	0 μg. per gm.	50 μg. per gm.	200 μg. per gm.	3000 μg. per gm.
1	3.2[1]	35.6	41.0	16.0
2	3.4	43.7	59.2	28.0
3	2.5	37.6	63.5	37.0
4	1.2	17.1	19.0	77.0

[1] These figures represent μg. aureomycin per gm. gut contents.

in the presence of penicillin. We have also observed (1954) a greater amount of pantothenic acid in the tract of rats receiving aureomycin and suboptimal amounts of pantothenic acid either orally or subcutaneously as compared with those animals which did not receive aureomycin.

Guggenheim *et al.* (1953) have also reported increased urinary and fecal excretion of certain B vitamins in the presence of several antibiotics.

Summary

A comparative study was made on the intestinal and cecal microbial population of 5-week-old pigs which had been raised from birth in the presence and absence of terramycin and whose gains had indicated a significant response to the antibiotic.

From the plate counts made of the organisms growing on four types of media including aerobic and anaerobic incubation, the data suggest that the effect of terramycin on the total numbers of microbes in all intestinal sections from the duodenum to the cecum is to increase their numbers about tenfold.

None of the various types of flora found in the gut of the basal animals was conspicuously absent from the contents of the gut of animals receiving the antibiotic.

Literature Cited

Braude, R., S. K. Kon and J. W. G. Porter. 1953. Antibiotics in nutrition. Nutr. Abstr. Rev. 23:473.

Grady, J. E. and W. L. Williams. 1952. Determination of aureomycin in feeds with the pad plate method. Lederle Laboratories, American Cyanamid Co., Pearl River, New York.

Guggenheim, K., S. Halevy, I. Hartmann and L. R. Zamir. 1953. The effect of antibiotics on the metabolism of certain B vitamins. Jour. Nutr. 50:245.

Johnson, B. Connor, M. F. James and J. L. Krider. 1948. Raising newborn pigs to weaning age on a synthetic diet with attempt to produce a pteroylglutamic acid deficiency. JOURNAL OF ANIMAL SCIENCE 7:486.

Nesheim, R. O., J. L. Krider and B. Connor Johnson. 1950. Antibiotics, whey and APF for baby pigs. JOURNAL OF ANIMAL SCIENCE 9:664.

Schendel, H. E. and B. Connor Johnson. 1953. Comparisons of anti-bacterial and anti-yeast-mold antibiotics with baby pigs in the presence of several levels of B vitamins. JOURNAL OF ANIMAL SCIENCE 12:88.

Schendel, H. E. and B. Connor Johnson. 1954. Studies of antibiotics in weanling rats administered suboptimal levels of certain B vitamins orally and parenterally. Jour. Nutr. In press.

Schendel, H. E. and B. Connor Johnson. 1954. Studies on antibiotic growth stimulation in weanling rats using radiothiamin as a tracer. Unpublished.

Wahlstrom, R. C., E. Cohn, S. W. Terrill and B. Connor Johnson. 1952. The effect of various antibiotics on baby pigs fed synthetic rations. JOURNAL OF ANIMAL SCIENCE 11:449.

Wahlstrom, R. C. and B. Connor Johnson. 1951. The effect of cortisone and of aureomycin on baby pigs fed a vitamin B_{12} deficient diet. Proc. Soc. Exptl. Biol. Med. 78:112.

Wahlstrom, R. C., S. W. Terrill and B. Connor Johnson. 1950. Effect of antibacterial agents on growth of baby pigs fed a synthetic diet. Proc. Soc. Exptl. Biol. Med. 75:710.

THE EFFECTS OF BACITRACIN, PENICILLIN AND ARSANILIC ACID ON GROWTH RATE AND FEED EFFICIENCY IN SWINE[1]

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STUDIES by a number of research workers during the past few years have shown that antibiotics added to swine rations have a growth promoting effect. The subject has been reviewed by Braude *et al.* (1953).

Substances besides the antibiotics have been suggested as a means of increasing feed efficiency and the growth rate of swine. The effects of arsanilic acid (*p*-amino phenylarsonic acid) in rations for swine have not been as spectacular as those of the antibiotics in increasing growth rate; however, arsanilic acid has been effective in increasing feed efficiency, general well being, and appearance. (Hanson *et al.*, 1953). Morehouse and Mayfield (1946) reported that 3-nitro-4-hydroxyphenyl arsonic acid when given in the drinking water at sub-coccidio-static levels had remarkable growth stimulating properties for chickens and turkeys. This finding prompted Morehouse (1949) to add arsenic compounds to the diet of chickens and turkeys. He observed that it increased growth rate and feed efficiency. This work was confirmed by Bird *et al.* (1949).

The question often arises whether arsenicals fed to swine will endanger the consumer of the meat. It has been ascertained that the deposition of arsenic in the chicken is not too high for human consumption of the meat when as much as 180 grams of arsanilic acid is added per ton of feed (Frost, 1953). Reports concerning the residual arsenic content of swine tissue are those of Carpenter (1951) and Hanson *et al.* (1953). Carpenter found 1.69 and 1.32 ppm of As_2O_3 in the liver and kidney, respectively, of swine fed 3-nitro-4-hydroxy phenylarsonic acid in their rations and slaughtered two hours after the last feeding. The length of the feeding period was 40 days. In contrast, Hanson reported that the amount of arsenic stored in the liver (per gram of

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fresh tissue) was approximately twice that in the kidney and muscle. After feeding 30, 60, and 90 grams of arsanilic acid per ton of feed—for a period from weaning (37 lb.) to market weight (200–210 lb.)—the content of arsenic in the liver was 2.74, 3.98 and 3.34 ppm of As_2O_3, respectively, as compared to 1.20, 2.04 and 2.19 ppm in the kidney. Fat and skin were low in arsenic at all levels of arsanilic acid fed.

From these considerations, it appeared that further studies on the effect of feeding an arsenical compound alone and in combination with various antibiotics would be desirable. The objectives of the following tests were: (1) to study the possible effects of bacitracin, arsanilic acid, penicillin, and a combination of bacitracin and penicillin and of bacitracin and arsanilic acid on growth rate and feed efficiency of swine; (2) to determine the amount of arsenic stored in the tissues of swine fed arsanilic acid; (3) to measure the loss of stored arsenic when the arsenical is withdrawn from the ration at different time intervals before slaughter.

Experimental Procedure

Six lots of seven Duroc pigs each were used in each of two experiments. In experiment 1, the pigs used were farrowed in concrete-floored pens where they remained for seven days before being placed with their dams on oat pasture. These pigs had access to a creep ration that contained an antibiotic from the time they were placed on pasture until they were brought to the swine center for use in this experiment. They were allotted on the basis of weight, sex, litter and condition. The pigs were fed in individual concrete-floored pens which were cleaned daily, and they had access to outside concrete pens. They were self-fed a 16-percent crude protein ration consisting of 77.0 percent ground milo, 16.8 percent soybean oil meal, 4.2 percent alfalfa leaf meal, 1.5 percent limestone, and 0.5 percent salt; the ration was fortified with riboflavin, 1.0 mg; niacin, 5.0 mg; pantothenic acid, 5.0 mg.; choline chloride, 250 mg.; and B_{12}, 7.5 mg. per pound of feed. All pigs were treated with sodium fluoride for round worms (Ascaris lumbricoides var. suis) before being placed on test.

In the second experiment, the pigs received no antibiotic before the beginning of the feeding trial. The initial weight of the pigs was about 20 pounds less than in experiment 1. They were alloted in the same manner and fed the same basal ration as in experiment 1; how-

TABLE 1. EFFECT OF BACITRACIN, PENICILLIN, AND ARSANILIC ACID ON FATTENING PIGS. EXPERIMENT NO. 1 [1]

Lot Number[2]	1	2	3	4	5	6
Treatment	Basal (Control)	10 gm. of Bacitracin per Ton	2 gm. Penicillin + 10 gm. Bacitracin per Ton	1 oz. Arsanilic acid per Ton	5 gm. Bacitracin + 1 oz. Arsanilic acid per Ton	10 gm. Bacitracin + 1 oz. Arsanilic acid per Ton
Av. initial wt., lb.	51.1	50.2	49.6	49.6	49.0	49.6
Av. final wt., lb.	192.4	191.1	210.7	198.0	199.3	199.4
Av. daily gain, lb.	1.68	1.68	1.92	1.77	1.79	1.78
Standard error	0.09	0.08	0.06	0.04	0.04	0.06
Feed/100 lb. gain, lb.	371	361	344	371	354	370

[1] The length of feeding period was 84 days, from December 16, 1952 to March 10, 1953. Bacitracin was used in the form of bacitracin methylene disalicylate (DPS).

[2] There were seven pigs per lot at the start of the experiment. One pig in lot 2 died of urinary calculi; one pig in lot 5 was removed from experiment owing to unthriftiness.

ever lot 5 in experiment 2 received penicillin alone, as indicated in table 2.

At the termination of the feeding trial in experiment 1, two pigs were slaughtered from the control lot and two from each of lots 4 and 6 (arsanilic acid and arsanilic acid plus bacitracin). In experiment 2, at the termination of the feeding trial, two pigs were slaughtered from the control lot and two from each of lots 4 and 6 (arsanilic acid and arsanilic acid plus bacitracin). Arsanilic acid was then removed from all rations, and two pigs from each of the different lots were slaughtered 5 days and 10 days later. The whole liver and both kidneys were taken immediately and frozen for later arsenic determination. After chilling

TABLE 2. EFFECTS OF BACITRACIN, PENICILLIN, AND ARSANILIC ACID ON FATTENING PIGS. EXPERIMENT NO. 2 [1]

Lot Number[2]	1	2	3	4	5	6
Treatment	Basal (Control)	10 gm. of Bacitracin per Ton	2 gm. Penicillin + 10 gm. Bacitracin per Ton	1 oz. Arsanilic acid per Ton	2 gm. Penicillin per Ton	10 gm. Bacitracin + 1 oz. Arsanilic acid per Ton
Av. initial wt., lb	31.4	31.4	31.4	31.4	31.4	31.3
Av. final wt., lb.	211.7	204.5	211.0	188.5	220.0	198.8
Av. daily gain, lb.	1.52	1.45	1.50	1.32	1.59	1.41
Standard error	0.04	0.07	0.09	0.05	0.04	0.04
Feed/100 lb. gain, lb.	367	350	340*	344*	334**	339*

[1] The length of the feeding period was 119 days, from July 14, 1953 to November 10, 1953.

[2] There were seven pigs per lot at the start of the experiment. One pig in lot 2 died of urinary obstruction; one pig in each of lots 5 and 6 was removed from the test because of unthriftiness.

* $P = .05$.

** $P = .01$.

the carcass for 24 to 48 hours, samples of muscle were taken from the posterior end of the loin and frozen for later arsenic determinations.

Having tried and discarded a number of different methods for the determination of small amounts of arsenic in animal tissues, the colorimetric method of Magnuson and Watson (1944) as modified by Maren (1946) was selected. Color was measured in the Beckman Spectrophotometer at 840 millimicrons wave length. The report of Sultzaberger (1943) that the peak of the adsorption curve is at 840 millimicrons was verified. In order to reduce sampling error, a relatively large sample (50 gm.) of tissue was ground in a Waring blender and 5-gram portions were then processed as outlined by Magnuson and Watson (1944). Before carrying out the arsenic determination on the tissue samples, the validity of the analytical procedure as carried out in this laboratory was established by recovery tests with As_2O_3 and As_2O_5 added to liver tissue. In a typical test, 20μg of As_2O_3 were added to three 5-gram samples of liver, each sample originally containing 0.60 mcg. of As_2O_3. Recoveries of total arsenic were 97.0, 97.0 and 96.7 percent.

Results and Discussion

All pigs in experiment 1 made excellent rates of gain (table 1). Although lot 3 (2 grams of penicillin plus 10 grams of bacitracin per ton of feed) gained 0.24 pound more per day than lot 1 (control), the difference between these two lots was not statistically significant, and this increase in gain was not repeated in the second experiment.

The results of experiment 2 are given in table 2. Small differences in growth rate between the controls and other lots were not statistically significant.

It should perhaps be pointed out that both experiments were conducted under excellent sanitary conditions which may account for the failure of the supplements to produce a marked increase in growth rate. These results are very similar to those obtained by Meade (1952) who observed a slight increase in rate of gain of pigs fed arsenicals singly or in combination with bacitracin.

A small but not statistically significant improvement in feed efficiency was observed in the first experiment with lots 2, 3, and 5, all of which received a bacitracin supplement (table1). In the second experiment, an improvement in feed efficiency was obtained with all lots fed an antibiotic, arsanilic acid or a combination of these (table 2). The

improvement in feed efficiency as compared to the control was statistically significant with the lots fed arsanilic acid alone, penicillin alone, a combination of arsanilic and bacitracin, and a combination of penicillin and bacitracin.

The results of the arsenic analyses are given in table 3. The content of As_2O_3 was several times greater in the liver than in the kidney or

TABLE 3. ARSENIC CONTENT OF LIVER, KIDNEY, AND MUSCLE FROM SWINE FED 1 OZ. ARSANILIC ACID PER TON OF FEED

Values expressed as p.p.m. of As_2O_3 in fresh tissue

Experiment	Withdrawal Time [1]	Liver [2]	Kidney [2]	Muscle [2]
	days			
1	0	4.6	1.30	1.00
2	0	5.20	1.50	1.30
2	5	0.75	0.70	0.71
2	10	1.00	0.90	0.50
Controls (no arsanilic acid fed)		0.12	0.08	0.08

[1] Days after withdrawal of arsanilic acid from feed before the pigs were slaughtered.
[2] Average values for four animals each, except that the values for the controls represent averages for 8 animals each.

muscle tissue. These data indicate that the accumulation of arsenic in the liver is somewhat greater in the pig than in the chicken. Arsenic rapidly disappeared from the liver after withdrawal of arsanilic acid from the feed. After 5 days the average content of As_2O_3 in the liver did not exceed 1 ppm.

During the slaughtering of the pigs, a visual examination of the liver, kidneys and spleen was made by the college veterinarians, but no abnormalities were observed.

It is of interest to compare the residual arsenic content of the swine tissue with the arsenic content of natural foods. Ellis *et al.* (1941) found 0.75 ppm of As_2O_3 in fresh-water fish and crustaceans. Holmes and Remington (1930) found from 1.5–4.1 ppm of As_2O_3 in cod, eels, and mackerel. Chapman (1926) and Coulson *et al.* (1935) reported shell fish and crustaceans to contain from 3–174 ppm As_2O_3.

Summary and Conclusion

Two experiments involving 84 weanling pigs are described in which bacitracin, penicillin and Arsanilic acid singly and in combination were added to a milo-soybean oil meal ration. None of these substances significantly increased the growth rate above the control in either of

the experiments. In the first experiment a small improvement in feed efficiency was obtained in the lots fed bacitracin alone, a combination of bacitracin and penicillin and a combination of bacitracin and arsanilic acid, but the differences did not prove to be statistically significant. In the second experiment, significant improvement in feed efficiency occurred in the lots fed arsanilic acid alone, penicillin alone, a combination of arsanilic acid and bacitracin, and a combination of penicillin and bacitracin.

The accumulation of arsenic in liver, kidney and muscle tissue after feeding arsanilic acid was greatest in the liver. On withdrawing arsanilic acid from the ration, the arsenic was rapidly lost from the liver, so that after 5 days the average content of As_2O_3 in the liver did not exceed 1 ppm.

Literature Cited

Bird, H. R., A. C. Groschke and M. Rubin. 1949. Effect of arsonic acid derivatives in stimulating growth of chickens. Jour. Nutr. 37:215.

Braude, R., H. D. Wallace and T. J. Cunha. 1953. The value of antibiotics in the nutrition of swine. A review. Antibiotics and Chemotheraphy, vol. III, No. 3.

Carpenter, L. E. 1951. The effect of 3-nitro-4-hydroxy phenyl arsonic acid on the growth of swine. Arch. Biochem. 32:181.

Chapman, C. A. 1926. On the presence of arsenic in marine crustaceans and shell fish. Analyst. 51:548.

Coulson, E. J., R. E. Remington and K. M. Lynch. 1935. Toxicity of naturally occurring arsenic in food. Jour. Nutr. 10:255.

Ellis, M. M., B. A. Westfall and M. D. Ellis. 1941. Arsenic in fresh-water fish. Ind. Eng. Chem. 33:1331.

Frost, D. V. 1953. Considerations on safety of arsanilic acid for use in poultry feeds. Poul. Sci. 32:217.

Hanson, L. E., L. E. Carpenter, W. J. Aunan, E. F. Ferrin and R. M. Anderson. 1953. The effect of various levels of arsanilic acid in the rations of market pigs. Minn. Agr. Exp. Sta. Bul. H110 (revised).

Holmes, A. D., and R. E. Remington. 1934. Arsenic content of American cod liver oil. Ind. Eng. Chem. 26:573.

Magnuson, H. J., and E. B. Watson. 1944. Microdetermination of arsenic in biological materials. Ind. Eng. Chem., Anal. Ed. 16:339.

Maren, T. H. 1946. Modification of the arsenic method of Magnuson and Watson. Ind. Eng. Chem., Anal. Ed. 18:521.

Meade, R. J. 1952. A study of antibiotics, combinations of antibiotics, arsanilic acid and an arsanilic acid bacitracin combination for growing fattening pigs in drylot. Neb. Agr. Exp. Sta. Prog. Rept. 307.

Morehouse, N. F. 1949. Accelerated growth in chickens and turkeys produced by 3-nitro-4-hydroxy phenyl arsonic acid. Poul. Sci. 28:375.

Morehouse, N. F. and O. J. Mayfield. 1946. The effect of some aryl arsonic acids on experimental coccidiosis infection in chickens. Jour. Parasitology. 32:20.

Sultzaberger, J. A. 1943. Microdetermination of arsenic in biological material. Ind. Eng. Chem., Anal. Ed. 15:408.

THE EFFECT OF PENICILLIN AND B-VITAMINS ON THE GROWTH OF PIGS FED DIFFERENT LEVELS OF PROTEIN [1, 2]

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EXCELLENT growth has been obtained with semi-purified diets in which natural protein sources were used, with pigs fed rations containing as low as 11 percent protein (Geurin *et al.*, 1951). Several recent studies have reported that less protein is required than formerly recommended when the ration is fortified with antibiotic and vitamins of the B-complex groups (Burnside *et al.*, 1951; Catron *et al.*, 1952 and Hoefer *et al.*, 1952). Hanson and Ferrin (1952) reported no benefit from adding riboflavin, niacin and calcium pantothenate to swine rations. However, Tribble *et al.* (1952) obtained a growth response in weanling pigs fed these three vitamins.

The different results obtained by various workers with vitamin supplementations may have been due to a varying content of B-vitamins in the basal rations. It is entirely possible that the increased rate of gain obtained on high protein rations in past years was due to the presence of an increased content of B-vitamins in the protein supplement and not due to an increased amino acid content of such rations. The possible interrelationships between vitamins, antibiotics and protien must also be considered. The present study was conducted to determine the effects of supplementing a mixed plant and animal protein ration with antibiotic (penicillin) and B-vitamins on the protein requirements of growing-fattening pigs in concrete drylot.

Experimental Procedure

Experiment 1

Forty-eight purebred weanling pigs averaging 43 to 44 lb. were allotted from outcome groups into four lots of twelve pigs each. Where outcome groups could not be made up of littermates, other comparable pigs were selected. The pigs were housed in concrete pens during the

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experiment. All pigs were fed and watered *ad libitum* and they were weighed bi-weekly. Each pig was removed from the experiment as it reached 200 lb. or after 119 days on the experiment.

The four treatments used consisted of two levels of protein (18 and and 14 percent) with and without an antibiotic-B vitamin supplement. When the pigs reached an average weight of approximately 130 lb. the protein level was reduced to 16 and 12 percent respectively. Penicillin was fed at a level of 5 mg. per lb. of ration and the B-vitamins included 6 mg. niacin, 5 mg. pantothenic acid, 1 mg. riboflavin, 4 mcg. vitamin B_{12} and 250 mg. choline chloride per lb. of ration.

The experimental rations were composed of ground yellow corn, solvent soybean oil meal and 60 percent digester tankage. A mineral mixture of ground limestone 2 parts, steamed bonemeal 2 parts and trace mineralized-salt 1 part was self-fed to all lots. The soybean meal and tankage were mixed at a 2:1 ratio in all rations and the various protein levels were obtained by adjusting the corn and soybean meal-tankage content of the ration.

Experiment II

Ninety-six purebred Duroc pigs which had been in continuous dry-lot with no previous antibiotic treatment were used in this experiment. They were allotted as equally as possible into 24 lots of four pigs each.

A replicated 3 x 4 factorial design was used. The three initial levels of protein studied were 18, 16 and 14 percent and the four treatments at each protein level were: control, B-vitamins, penicillin, and B-vitamins plus penicillin. The protein content of the ration was reduced four percentage units to 14, 12 and 10 percent, respectively when the pigs averaged 100 lb. Average initial weight of the lots was 30 lb. per pig.

The basic feeds and the antibiotic-B-vitamin supplement used in the rations were essentially the same as in the first experiment. However, in this study the trace mineralized salt and steamed bonemeal were included in the mixed ration. A vitamin A and D supplement was also added in order to furnish 1000 USP units of vitamin A and 125 USP units of Vitamin D per lb. of ration. The calculated calcium and phosphorus content of all the rations was held constant by adjusting the amount of steamed bonemeal.

Seventy-four animals were slaughtered at the finish of this experiment. Carcass measurements included back fat, body length and the cross section area of the longissimus dorsi muscle.

Experiment III

The third experiment was designed to study only differences in protein level on rations supplemented with B-vitamins and penicillin. The rations used were the same as in the previous experiment.

Thirty-two purebred Duroc pigs were alloted into eight lots of four pigs each. The four protein levels used were 18, 16, 14 and 12 percent from weaning to an average weight of 100 lb. and 14, 12, 10 and 8 percent respectively from 100 to 200 lb. Two lots of pigs received each level of protein.

TABLE 1. SUMMARY OF RESULTS FOR EXPERIMENT I

Protein level, percent	18–16	18–16	14–12	14–12
Antibiotic-Vitamin supplement	+	—	—	+
Number of pigs [1]	14	13	14	13
Days on experiment	94	115	119	92
Av. initial wt., lb.	43	44	43	44
Av. final wt., lb.	200	178	171	201
Av. daily gain, lb.	1.67**	1.17	1.07	1.70**
Av. daily feed per pig, lb.	5.8	4.5	4.3	5.8
Av. feed per pound gain, lb.	3.48	3.84	4.04	3.39

[1] One pig died in each of lots 2 and 4 from a non-nutritional cause.
** Significant at a probability of 0.01.

Results and Discussion

Experiment I

A summary of the results of the first experiment is presented in table 1. No significant differences in daily gains resulted by varying the protein level either in the presence or absence of the penicillin and B-vitamins. Penicillin-B vitamins supplementation had a highly significant effect on rate of gain of the pigs on either level of protein. The response obtained was of similar magnitude during both early growth (initial to 130 lb.) and the later period (130 to 200 lb.).

The data on feed required per 100 lb. of gain show a considerable saving in feed when the rations were fortified with penicillin and B-vitamins. The pigs on the higher protein ration required 9.4 percent less feed per unit of gain and those which received the lower protein ration required 16.1 percent less feed than the pigs on these same rations without penicillin and B-vitamins. This difference would very likely have been greater had all lots been fed to 200 lb. live weight.

TABLE 2. SUMMARY OF AVERAGE DAILY GAINS BY WEIGHT PERIODS FOR EXPERIMENT II[1] (lb.)

Treatment	Control	B Vitamins	Penicillin	B-Vitamins and Penicillin	Av.
Initial to 100 lb.					
18 percent protein	1.10	1.24	1.32	1.42	1.27
16 " "	1.11	1.23	1.23	1.41	1.25
14 " "	1.07	1.20	1.21	1.31	1.20
Average	1.09	1.22	1.25	1.38	
100 to 200 lb.					
14 percent protein	1.84	2.05	2.10	2.10	2.02
12 " "	1.86	2.00	2.00	2.17	2.01
10 " "	1.66	1.98	1.80[2]	2.04	1.87
Average	1.79	2.01	1.98	2.10	
Initial to 200 lb.					
18–14 percent	1.43	1.62	1.70	1.76	1.62
16–12 "	1.46	1.59	1.60	1.78	1.59
14–10 "	1.33	1.60	1.49	1.67	1.51
Average	1.40	1.60	1.59	1.74	

[1] Two lots of four pigs each per treatment.
[2] The gain of two pigs which died from poisoning was estimated (Snedecor 1946).

Experiment II

The average daily gains and feed per 100 lb. gain are shown in tables 2 and 3. The analysis of variance of average daily gains is presented in table 4.

A significantly greater rate of gain was shown by the pigs receiving either the five B-vitamins or the penicillin during both the initial and final growth phases. The addition of one or the other of these two supplements to the diet did not produce a maximum rate of gain however, particularly during the period from weaning to 100 lb., since the inclusion of both penicillin and the B-vitamins in the ration induced

TABLE 3. SUMMARY OF FEED REQUIRED PER 100 POUND GAIN FOR EXPERIMENT II[1] (lb.)

Protein Levels (%)	18–14	16–12	14–10
Control	340	333	336
B-vitamins	348	316	335
Penicillin	350	328	337
B-vitamins and penicillin	309	302	353

[1] Two lots of four pigs each per treatment.

greater weight gains than either supplement alone. In most cases the two supplements were essentially additive in their effects on rate of gain when given simultaneously—an observation supported by low F values for interaction of penicillin and B-vitamins (table 4).

There was no significant difference in average daily gains made by the pigs on the different protein levels during the period up to 100 lb.

TABLE 4. ANALYSIS OF VARIANCE OF AVERAGE DAILY GAINS FROM WEANING TO 200 LB. FOR EXPERIMENT II

Source of Variation	d. f.	Mean Square	F Value
Total	93	....	
Replicates (Rep)	1	.174	
B-vitamins (Vit)	1	.662	27.7**
Penicillin (Pen)	1	.662	27.7**
Protein levels	2	.091	3.8*
Protein x Rep	2	.016	
Protein x Vit	2	.016	
Protein x Pen	2	.019	
Vit x Rep	1	.008	
Vit x Pen	1	.020	
Pen x Rep	1	.017	
Protein x Rep x Vit	2	.018	
Protein x Rep x Pen	2	.004	
Protein x Vit x Pen	2	.015	
Vit x Rep x Pen	1	.001	
Protein x Rep x Vit x Pen	2	.002	
Error	70	.024	

* Significant at a probability of 0.05.
** Significant at a probability of 0.01.

However, during the period from 100 to 200 lb. the pigs receiving the 10 percent protein ration without the five B-vitamins made considerably slower gains than did those pigs receiving similar rations of 12 or 14 percent protein. It would appear from this experiment that the 10 percent protein ration was limiting more in one, or more, of the five B-vitamins fed than in amount or quality of protein. When the data are summarized for the entire growth period a significant difference is noted between protein levels (table 4).

Feed required per 100 lb. of gain was quite variable between groups. It was noted that some lots persisted in wasting more feed than others and since no attempt was made to determine wastage this may account for some of the variation. All lots were quite efficient with the high and medium protein lots supplemented with both peni-

cillin and B-vitamins being particularly efficient in feed conversion. The pigs receiving the unsupplemented rations consistently consumed less feed per day than did those pigs receiving the B-vitamins or penicillin either alone or in combination.

The results of the carcass studies are presented in table 5. No significant differences existed between backfat thickness or carcass length,

TABLE 5. SUMMARY OF CARCASS MEASUREMENTS FOR EXPERIMENT II

Item	No. Pigs	Av. Slaughter Wt., Lb.	Av.[1] Carcass Length (inches)	Av.[2] Back Fat (inches)	Av.[3] Loin Lean Area (square inches)
Protein Levels (%)					
18–14	25	199	29.1	1.75	3.57*
16–12	26	202	29.1	1.78	3.63**
14–10	23	200	28.9	1.85	3.11
Treatments					
Control	18	202	29.2	1.74	3.28
B-vitamins	18	200	29.1	1.78	3.62*
Penicillin	20	199	28.9	1.80	3.14
B-vitamins and penicillin	18	200	29.0	1.83	3.79**

[1] Length measured from the first rib to the point of the aitch-bone.
[2] Average of four measurements taken at first, seventh, and last rib and last lumbar vertebrae.
[3] Product of length times width of longissimus dorsi muscle cut between 6th and 7th ribs from the loin end.
* Significant at a probability of 0.05.
** Significant at a probability of 0.01.

although the pigs fed the low protein rations produced carcasses with slightly more backfat than those fed the high or medium protein rations. A significant difference was found in the loin lean area with the low protein group having the smaller loin eye muscle. This difference was also noted between treatments where the addition of B-vitamins, alone or in combination with penicillin, significantly increased the loin lean area.

If the loin lean area is used as a measure of the leanness of the carcass these results are in general agreement with the findings of Wilson *et al.* (1953). These authors reported that supplements of aureomycin and vitamin B_{12}, alone or in combination, increased the percentage of lean in the carcass. They also found that pigs receiving high protein rations produced leaner carcasses than those fed low protein rations.

Experiment III

The third experiment is summarized in table 6. The average daily gains made by the pigs on the three higher levels of protein were essentially the same during the period from weaning to 100 lb. A deficiency of protein in the 12 percent protein ration is suggested by the poorer growth during this initial period.

TABLE 6. SUMMARY OF GROWTH AND FEED EFFICIENCY DATA FOR EXPERIMENT III[1]

Protein level (%)	18–14	16–12	14–10	12–9
		Average daily gains (lb.)[2]		
Weaning to 100 lbs.	1.44	1.38	1.38	1.19
100 to 200 lbs.	2.24	2.28	2.15	1.25
Weaning to 200 lbs.	1.82	1.80	1.74	1.22
		Average daily feed (lb.)		
Weaning to 100 lbs.	3.9	3.8	4.0	3.8
100 to 200 lbs.	8.9	8.4	8.3	5.8
Weaning to 200 lbs.	6.3	6.0	6.0	4.8
		Feed consumed per 100 pounds gain (lb.)		
Weaning to 100 lbs.	272	275	290	315
100 to 200 lbs.	396	368	387	469
Weaning to 200 lbs.	344	331	347	391

[1] Two lots of 4 pigs each per treatment.
[2] Average initial weight 31.0 lb.

Similar results were obtained during the period from 100 to 200 lb. body weight. As in the previous experiment there was a slight reduction in rate of gain made by the pigs on the 10 percent protein level during this period. However, the greatest difference was shown by the very poor growth of the pigs receiving the 9 percent protein ration. This ration did not contain any protein supplement but was composed of ground corn and minerals and fortified with the B-vitamin-antibiotic supplement used in these studies. There was more variation in rate of gain in the lots fed this ration than in any of the other lots.

An improvement in efficiency of feed utilization is apparent at the higher levels of protein. There was a steady increase in feed efficiency as the percent protein was increased during the period up to 100 lb. This did not occur in the period from 100 to 200 lb. Feed efficiency

was quite variable during this period with the low protein group again requiring the most feed per unit of gain.

Summary

Three experiments were conducted to study the effect of penicillin and B-vitamins on the growth of pigs fed different levels of protein.

Penicillin at 5 mg. per lb. of total ration and/or B-vitamin supplementation (riboflavin, niacin, pantothenic acid, vitamin B_{12} and choline) had a highly significant effect in improving rate of gain. This response was shown at all levels of protein fed.

The data indicate that the 10 percent protein ration was limiting in one or more of the five B-vitamins fed more than in amount or quality of protein when fed to pigs over 100 lb. in weight. A 12 percent protein ration produced slower gains than higher levels of protein from weaning to 100 lb. and a 9 percent protein ration did not support normal growth from 100 lb. to market weight.

Significantly leaner carcasses, as measured by loin-lean area, were produced by pigs receiving the B-vitamin supplementation or the high and medium protein rations as compared to those getting no B-vitamin supplement or the low protein ration respectively.

Very little difference in rate of gain or feed efficiency was obtained between the 18–14, 16–12 or 14–10 percent protein levels when fed in corn-soybean meal-tankage rations supplemented with minerals, vitamins and penicillin. However, the results suggest the medium protein combination would be preferred from an over-all economic viewpoint due to the lowering of carcass quality in the low protein group.

Literature Cited

Burnside, J. E., R. H. Grummer, P. H. Phillips and G. Bohstedt. 1951. The effect of vitamin B_{12} and aureomycin when fed with high, intermediate and low protein levels to swine. JOURNAL OF ANIMAL SCIENCE 10:1042.

Catron, D. V., A. H. Jensen, P. G. Homeyer, H. M. Maddock and G. C. Ashton. 1951. Re-evaluation of protein requirements of growing-fattening swine as influenced by feeding an antibiotic. JOURNAL OF ANIMAL SCIENCE 11:221

Geurin, H. B., J. A. Hoefer and W. M. Beeson. 1951. Protein efficiency of corn by-products and other protein feeds for swine growth. JOURNAL OF ANIMAL SCIENCE 10:607.

Hanson, L. E. and E. F. Ferrin. 1952. Riboflavin, niacin, and calcium pantothenate as supplements to a mixed ration for growing pigs. JOURNAL OF ANIMAL SCIENCE 11:762.

Hoefer, J. A., R. W. Luecke, F. Thorp, Jr. and R. L. Johnston. 1952. The effect of terramycin on the growth of pigs fed different levels of protein. JOURNAL OF ANIMAL SCIENCE 11:455.

Snedecor, G. W. 1946. Statistical Methods. 4th Ed. Iowa State College Press, Ames, Iowa.

Tribble, L. F., J. F. Lasley and A. G. Hogan. 1952. B vitamin deficiencies in rations for weanling pigs. JOURNAL OF ANIMAL SCIENCE 11:778.

Wilson, G. D., J. E. Burnside, R. W. Bray, P. H. Phillips and R. H. Grummer. 1953. Pork carcass value as affected by protein level and supplementation with aureomycin and vitamin B_{12}. JOURNAL OF ANIMAL SCIENCE 12:291.

QUANTITATIVE PHENYLALANINE REQUIREMENT OF THE WEANLING PIG [1]

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QUANTITATIVE requirements for a number of essential amino acids needed by the growing pig have been published. Investigators at Purdue and Cornell have reported the quantitative needs for tryptophan (Shelton *et al.*, 1951a), lysine (Brinegar *et al.*, 1950a; Shelton *et al.*, 1951b), methionine (Shelton *et al.*, 1951c; Curtin *et al.*, 1952a), isoleucine (Brinegar *et al.*, 1950b), threonine (Beeson *et al.*, 1953), and valine (Jackson *et al.*, 1953). The phenylalanine requirement is reported herein. Currently, only the quantitative requirements for histidine, leucine, and arginine remain to be elucidated; the essential nature of these amino acids has been shown (Loosli *et al.*, 1951; Mertz *et al.*, 1952). Mertz *et al.* (1952) used purified diets containing only nine essential amino acids without phenylalanine. Pigs fed such rations gained only 0.00 to 0.04 lb. daily. When 0.9 percent DL-phenylalanine was added, the daily gain was 1.12 to 1.29 lb. This established the essential nature but not the requirement for phenylalanine.

Diets containing proteins or protein hydrolysates deficient in certain amino acids have been used previously to determine quantitative amino acid requirements. The threonine (Beeson *et al.*, 1953) and valine (Jackson *et al.*, 1953) requirements were determined by employing diets which included large amounts of corn. Appropriate quantities of the ten essential amino acids and diammonium citrate were added to provide protein of sufficient quality and quantity. In the present trial varied quantities of DL-phenylalanine were added to a similar diet, low in phenylalanine and tyrosine, in order to determine the amount sufficient for optimum response.

Experimental

The major components of the basal diet (table 1) were corn, cerelose, nine essential amino acids, minerals, diammonium citrate, and liver

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extract. Necessary vitamin supplements were mixed with the daily feed and sufficient amounts of terramycin and vitamin B_{12} were included in the ration. The basal diet contained 12.60 percent total crude protein, 6.01 percent being supplied by corn, 4.93 percent by amino acids, and 1.66 percent by diammonium citrate. In the other diets, different amounts of DL-phenylalanine replaced portions of cerelose and diammonium citrate, so that the protein level remained constant. In previous tests (Beeson *et al.*, 1953; Jackson *et al.*, 1953) a level of 13 percent crude protein produced normal growth in weanling pigs when adequate levels of the ten essential amino acids were fed.

TABLE 1. MAJOR COMPONENTS OF BASAL RATION (12.6% CRUDE PROTEIN)

Ingredient	Percent
Ground corn [1]	66.00
Cerelose	21.57
Nine essential amino acids [2]	5.48
Minerals [3]	4.30
Diammonium citrate	2.15
Liver extract	0.50
	100.00 [4]

[1] Contained 9.1 percent crude protein and 13.8 percent moisture by analysis (A.O.A.C., 1945).
[2] See table 2.
[3] Beeson *et al.* (1953).
[4] Crystalline terramycin·HCl (10 mg. per lb. feed) and crystalline B_{12} (10 mcg. per lb. feed) were added to all diets. Vitamin supplements used by Mertz *et al.* (1952) were fed daily.

The amino acid composition of the corn [1] was determined by the microbiological method of Steele et al. (1949) and is presented in table 2 along with the total amino acid content of the basal diet. With the exception of phenylalanine, this ration provides sufficient quantities of utilizable amino acids (Jackson *et al.*, 1953) to support efficient growth. The basal ration contained 0.23 percent phenylalanine and 0.14 percent tyrosine. Since tyrosine can replace about one-half of the phenylalanine needed in the diet of the rat (Womack and Rose, 1946), it was assumed that the 0.14 percent tyrosine in the basal ration was equivalent to the same amount of phenylalanine, and that the ration is equivalent for growth purposes to one containing 0.37 percent phenylalanine.

[1] The authors are indebted to Mrs. Dominic Colasito and Mr. Ricardo Bressani for these analyses.

TABLE 2. AMINO ACID COMPONENTS OF THE BASAL DIET USED TO DETERMINE THE PHENYLALANINE REQUIREMENT OF THE WEANLING PIG

Amino Acid	Amino Acid Assay Value of Corn	Digestible Amino Acid in the Diet Supplied by Corn [1]	Added Crystalline Amino Acid in the Diet	Form of Amino Acid Used	Total Utilizable Amino Acid [2] In Diet		In the Digestible Crude Protein of Diet [3]	
	%	%	%		%		%	
Arginine	0.29	0.16	0.05	L·HCl	0.20		1.72	(1.6)
Histidine	0.20	0.11	0.39	L·HCl·H_2O	0.40		3.45	(3.2)
Isoleucine	0.29	0.16	1.08	L with D-allo	0.70		6.03	(5.6)
Leucine	1.08	0.60	0.20	L	0.80		6.90	(6.4)
Lysine	0.18	0.10	2.25	DL·HCl	1.00		8.62	(7.9)
Methionine	0.16	0.09	0.51	DL	0.60		5.17	(4.8)
Phenylalanine	0.41	0.23	0.00	DL	0.23	0.37 [4]	1.98	(1.8)
Tyrosine	0.26	0.14	0.00		0.14		1.21	(1.1)
Threonine	0.28	0.15	0.49	DL	0.40		3.45	(3.2)
Tryptophan	0.04	0.02	0.18	DL	0.20		1.72	(1.6)
Valine	0.42	0.23	0.33	DL	0.40		3.45	(3.2)

[1] Assay value x percent of corn in diet x percent digestibility of corn protein, or assay value x 66 percent x 84 percent.
[2] Digestible amino acid supplied by corn plus physiologically active amino acid from the crystalline form.
[3] Digestible crude protein = 11.6 percent of ration. Figures in parentheses denote percent of utili.able amino acid in the total crude protein of the diet. Total crude protein = 12.6 percent of ration.
[4] At lowest level of phenylalanine.

Twelve weanling Duroc male pigs (barrows) averaging 25 lb. were divided equally into four lots according to litter origin and weight. Each pig was fed individually *ad libitum* twice daily. Vitamin supplements which met the daily vitamin needs of each pig were added to the evening feed. Sufficient amounts of DL-phenylalanine were added to the basal ration to provide 0.37, 0.46, 0.56 and 0.66 percent total digestible combination of phenylalanine plus 0.14 percent tyrosine in the experimental treatments. The pigs were fed for 28 days.

TABLE 3. AVERAGE DATA[1] FOR PIGS RECEIVING VARIOUS LEVELS OF PHENYLALANINE IN THE DIET

Added DL-phenylalanine, %	0.0	0.09	0.19	0.29
Total utilizable phenylalanine, + tyrosine,[2] %	0.37	0.46	0.56	0.66
Initial weight (lb.)	25.6	25.3	24.9	25.7
Final weight (lb.)	40.9	49.1	47.3	47.1
Daily gain (lb.)	0.55	0.85	0.80	0.76
Daily feed consumption (lb.)	1.79	2.34	2.22	2.30
Feed per pound of gain (lb.)	3.26	2.75	2.78	3.01
Protein efficiency[3]	2.45	2.90	2.87	2.66

[1] Three male pigs per treatment. Duration of experiment 28 days.
[2] Includes 0.14 percent tyrosine.
[3] Pounds gain per pound crude protein.

Results and Discussion

Rose and Womack (1946) reported that D-phenylalanine in the diet of rats induces almost as satisfactory growth as the natural isomer. Nasset and Siliciano (1952) found the racemic mixture to be as effective as the natural form of phenylalanine for maintenance of nitrogen equilibrium in the adult rat. Therefore, we have assumed that both isomers in DL-phenylalanine are effectively utilized by the pig, and no distinction will be made between isomers in expressing the phenylalanine requirement of the pig.

The effects of different levels of phenylalanine upon growth, appetite, and feed efficiency are illustrated in table 3. Although the 0.37 percent total digestible combination of 0.23 percent phenylalanine plus 0.14 percent tyrosine was inadequate for optimum growth, 0.46 percent of these combined amino acids supported more rapid growth than either 0.56 or 0.66 percent. Further, appetite and feed conversion data indicated superiority of response when the 0.46 percent level was fed. Although pigs which received the 0.56 percent level (0.42 percent phenylalanine

plus 0.14 percent tyrosine) gained almost as efficiently as those which were fed the 0.46 percent level, they consumed less feed and consequently gained at a slower rate.

Maximum gain and feed utilization were realized by feeding a diet which contained 0.46 percent total digestible phenylalanine plus tyrosine. Therefore, the requirement of the weanling pig for phenylalanine (assuming 0.14 percent tyrosine replaces 0.14 percent phenylalanine) is suggested to be 0.46 percent of the ration. Such a level is approximately one-half of the requirement of the rat. (0.9 percent) as reported by Rose *et al.* (1946) using mixtures of purified amino acids. However, the requirement of the rat is reduced to that of the pig when diets containing corn are fed (Mertz *et al.*, 1954).

Summary

Semi-purified diets containing corn, dextrose, vitamins, minerals, and amino acids were used to determine the phenylalanine requirement of the weanling pig. The basal ration contained 0.37 percent total digestible aromatic amino acids (0.23 percent phenylalanine plus 0.14 percent tyrosine) from corn. Other rations included sufficient DL-phenylalanine to supply 0.46, 0.56, and 0.66 percent of these two amino acids in the diet, the tyrosine level remaining at 0.14 percent in all cases.

Maximum gains and feed utilization were obtained with 0.32 percent phenylalanine and 0.14 percent tyrosine in the ration. Assuming that the 0.14 percent tyrosine substituted for an equal weight of phenylalanine, the phenylalanine requirement of the weanling pig is 0.46 percent of the ration. This value is equivalent to 4.0 percent of the digestible crude protein or 3.6 percent of the total protein in the diet.

Literature Cited

Association of Official Agricultural Chemists. 1945. Methods of Analysis. A.O.A.C., Washington, D. C.

Beeson, W. M., H. D. Jackson and E. T. Mertz. 1953. Quantitative threonine requirement of the weanling pig. JOURNAL OF ANIMAL SCIENCE 12:870.

Brinegar, M. J., H. H. Williams, F. H. Ferris, J. K. Loosli and L. A. Maynard. 1950a. The lysine requirement for the growth of swine. Jour. Nutr. 42:129.

Brinegar, M. J., J. K. Loosli, L. A. Maynard and H. H. Williams. 1950b. The isoleucine requirement for the growth of swine. Jour. Nutr. 42:619.

Curtin, L. V., J. K. Loosli, J. Abraham, H. H. Williams and L. A. Maynard. 1952a. The methionine requirement for the growth of swine. Jour. Nutr. 48:499.

Jackson, H. D., E. T. Mertz and W. M. Beeson. 1953. Quantitative valine requirement of the weanling pig. Jour. Nutr. 51:109.

Loosli, J. K. 1951. Amino acid requirements of swine. Soybean Digest 11: No. 6, 26

Mertz, E. T., W. M. Beeson and H. D. Jackson. 1952. Classification of essential amino acids for the weanling pig. Arch. Biochem. Biophys. 38:121.

Mertz, E. T., R. Bressani and H. D. Jackson. 1954. Unpublished data. Purdue University. Lafayette, Indiana.

Nasset, E. S. and A. M. Siliciano. 1952. Nitrogen balance of adult rats fed diets low in L-, DL-, and D-phenylalanine. Jour. Nutr. 47:437.

Rose, W. C. and M. Womack. 1946. The utilization of the optical isomers of phenylalanine and the phenylalanine requirement for growth. Jour. Biol. Chem. 166:103.

Shelton, D. C., W. M. Beeson and E. T. Mertz. 1951a. Quantitative DL-tryptophan requirement of the weanling pig. JOURNAL OF ANIMAL SCIENCE 10:73.

Shelton, D. C., W. M. Beeson and E. T. Mertz. 1951b. Quantitative L-lysine requirement of the weanling pig. Arch. Biochem. Biophys. 30:1.

Shelton, D. C., W. M. Beeson and E. T. Mertz. 1951c. The effect of methionine and cystine on the growth of weanling pigs. JOURNAL OF ANIMAL SCIENCE 10:57.

Steele, B. F., H. E. Sauberlich, M. S. Reynolds and C. A. Baumann. 1949. Media for Leuconostoc Mesenteroides P-60 and Leuconostoc Citrovorum 8081. Jour. Biol. Chem. 177:533.

Womack, M. and Rose, W. C. 1946. The partial replacement of dietary phenylalanine by tyrosine for purposes of growth. Jour. Biol. Chem. 166:429.

SALT POISONING OF PIGS[1]

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LOSSES from salt poisoning of pigs seem to have occurred during many generations of stockmen. In letters or other reports of swine feeders, discarded meat brine and salty whey from cheese factories, constituting perhaps the last drippings into an empty whey tank, have been held responsible for such cases of poisoning. A craving for salt may have played a part in causing an excessive intake of salt, especially when present in a palatable liquid. Management of afflicted pigs, the steps leading up to the occurrences of poisoning or therefore the conditions necessary for poisoning pigs, usually have not been specifically indicated and have been difficult to establish.

A question may be whether the term "poisoning" as applied to salt is appropriate, or whether it is merely a heavy concentration of salt in the digestive tract of pigs that may cause fatal dehydration of vital tissues. This is an academic question, however, for in the end, whatever the specific effect of large amounts of salt, a pig is dead.

Previous Attempts at Salt Poisoning

In view of the fairly frequent occurrence of such losses, and with the desire to be able to forestall them, the Wisconsin Experiment Station (Grummer and Bohstedt, 1947), several years ago attempted to bring about conditions of salt poisoning. The rations fed consisted of corn balanced with soybean oil meal and limestone, therefore ration mixtures that were low in sodium and chlorine. In these earlier experiments the pigs that in all cases were kept in dry lot, were self-fed such mixtures for months at a time. Thereupon they were exposed to either large amounts of dry salt or, after a few weeks of slop feeding containing amounts of salt varying from zero to 1.5 percent, were then exposed to slop containing 2 percent salt on the wet basis.

None of the less extreme amounts of salt in the slop or swill in the fore part of the tests had proved noticeably unpalatable or injurious. When thereupon all lots were exposed to 2 percent salt

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in the slop, the pigs consumed it sparingly at first, and after 12 hours left it untouched. None of them died. Similar negative results with salt starvation followed by sudden salt surfeit were obtained by the Indiana Agricultural Experiment Station (Vestal, 1947).

A search in the literature on salt in relation to swine management has not revealed any successful experimental attempt at salt poisoning pigs through feeding, or through salt starvation and subsequent exposure to large amounts of salt. These failures in apparently all attempts at salt poisoning pigs set us thinking that perhaps feeding practices at agricultural colleges and experiment stations were not sufficiently comparable to those existing on many farms, especially in reference to competition of slop- or swill-fed pigs at feed troughs. Thus the animals in their excitement and greed might bolt an overdose of salt in their otherwise customary feed. A previous salt deficiency in their rations might accentuate the rate of consumption and poisoning effect. An increasingly restricted trough space in its turn might step up a spirit of competition for feed at the trough. Lack of free access to water under such conditions might be another contributing factor to salt poisoning.

Method

Two lots of six pigs each, averaging 65 lbs. apiece, were hand-fed swill twice a day, one group full fed and the other group limited to approximately 80 percent of the feed intake of the first group. The feed mixture in every case was allowed to soak 24 hours or longer, using three parts of water by weight to one part of the feed mixture, which consisted of the following: Ground yellow corn 57.25 lb. wheat flour middlings 28.00 lb., linseed meal 14.00 lb., ground limestone 0.75 lb., antibiotic feed supplement 0.50 lb., totaling 100.5 lb.

The length of trough was purposely limited so that while adequate during the early weeks of the experiment, nevertheless with increasing size of the pigs it proved too short. This situation was conducive to stimulating competition for feed. Additional water was freely accessible to both groups. During the early part of the experiment bloody scours affected both lots and caused the death of two animals in the limited fed group. Thereafter all the animals made uniform gains, slightly below normal. About 2 months after the start of the experiment the pigs showed a craving for salt by licking urine from the concrete floor and by chewing wood.

Where the experiment was started July 25, 1951, 99 days later, on November 1, Thursday, the regular morning feed was omitted and the drinking water removed. At 1:00 p.m. the full-fed group of six pigs was fed a swill that contained 1.5 percent salt on the actual wet basis, or 6.0 percent on the air-dry feed basis. The six pigs consumed approximately 60 lb. of the feed for that day. Each pig therefore consumed about 2.4 ounces of salt in one day.

The limited-fed group, now consisting of four pigs, was given approximately 40 lb. of a 2-percent-salt swill. This therefore contained 8.0 percent salt on the air-dry feed basis, giving each pig about 3.2 ounces of salt that day. In neither case was any additional water allowed.

Both lots cleaned up the feed although the group getting the 2-percent-salt swill acted more hesitant about eating than those on the lower salt level.

Results

No visible symptoms were apparent by noon of the following day, Friday, where meanwhile both lots had been without feed and water. At 1:00 o'clock that day, or 24 hours since the last feeding, both lots were given a 2-percent-salt swill in amounts of approximately 10 lb. per head. Both groups consumed the feed reluctantly. By 6:00 o'clock that evening they had not finished their feed. They showed no signs of distress even though no additional water was available to them. At this time it seemed as if even by this system of feeding, which was intended to approximate conditions that might exist on some farms, pigs could not be poisoned by sudden excessive intakes of salt.

But at 7:00 o'clock on the morning of November 3, Saturday, three pigs were found in a paralyzed semiconscious state. One pig lying on its side and pawing convulsively on the concrete floor, bled profusely from the resulting leg abrasions. This animal died before 10:30 a.m., and all three animals were posted by Dr. Dale K. Sorensen [3] between 10:30 and 11:30 that same forenoon. By 6:00 p.m. two more animals in the full-fed lot were down. Other animals in both lots exhibited distress, having a staggering gait. None of the pigs in either lot was affected by scours as a result of heavy salt feeding. Both lots meanwhile had been given plain drinking water and normal swill.

On Sunday, November 4, one animal in the limited-fed lot died, and another one was down. The two animals that were down in the full-fed lot the previous day were still alive.

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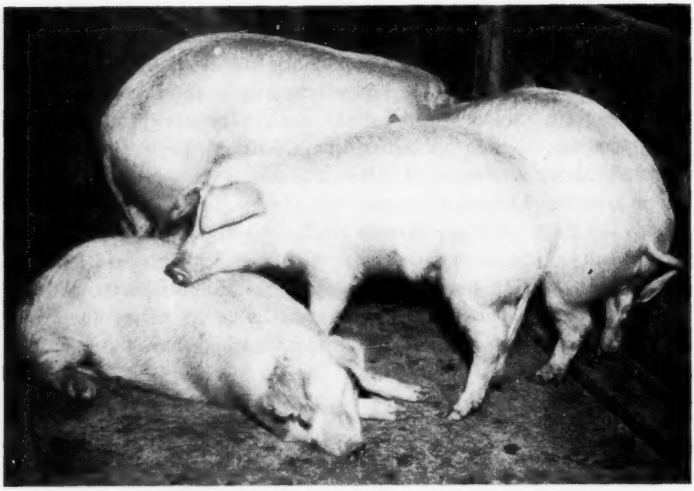

Figure 1. Pigs beginning to show signs of salt poisoning.

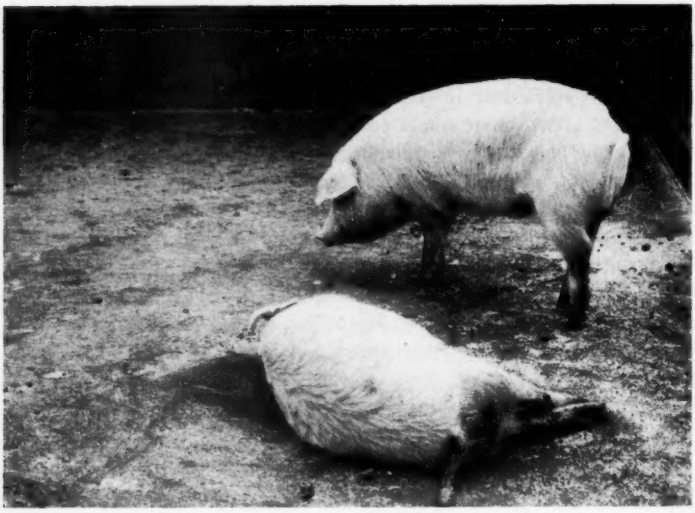

Figure 2. Pigs in advanced stages of salt poisoning. Three are down and are exhibiting clonic spasms of legs.

On Monday, November 5, the three paralyzed animals were posted by Dr. Sorensen. This left one in the full-fed and two in the limited-fed lots that were transferred to another barn and fed normal feed and water. The following day one of these pigs was discovered to be blind. But all three pigs fed normal rations at this time and subsequently, made good gains.

Autopsy

The autopsy report on animals which were sacrificed on this salt feeding experiment was as follows:

Symptoms: The most marked symptoms were the nervous symptoms, staggering, marked weakness, paresis and paralysis of the hind parts or general paralysis. Blindness was observed in one animal, which did not succumb and was taken off the salt ration.

External Appearance: The first three animals autopsied on 11/3/51 were comatose in a lateral recumbent position. Clonic spasms of the legs were very evident and abrasions on the dependent portion of the legs existed from contact with the cement floor. The animals were covered with fresh unclotted blood, which apparently came from these abrasions. The other two animals sacrificed on 11/5/51 had abrasions but no free blood was observed.

Heart: Slight increase in the amount of fluid in the pericardial sac. Numerous subendocardial hemorrhages were observed but they were attributed to agonal struggles as a result of sacrificing the animals by exsanguination. The hearts were flabby and atonic.

Lungs: No visible lesions.

Liver: The livers of all animals were extremely pale and yellow in color suggesting anemia and fatty changes. They were very friable and a few ecchymotic hemorrhages were observed beneath the capsule.

Spleen: The spleens on four animals were apparently normal. The fifth one had many recent red infarcts along the free borders.

Kidneys: On one animal, the kidneys were smaller and firmer than usual. The rest were apparently normal in size, color, and consistency. The medullary portion may have been smaller than normal.

Adrenals: No gross changes observed.

Bladder: No visible lesions.

Genitalia: No visible lesions.

Stomach: No visible lesions.

Intestine: Few ascarids were seen in the duodenum of all animals. Few areas of ecchymotic hemorrhages in the jejunum and the colon was pinkish in color.

Lymph Nodes: Apparently normal.

Brain: Apparently normal.

Gross Anatomic Diagnosis: From the history and clinical symptoms the cause of the condition was undoubtedly due to the toxicity of the NaCl or the stress exerted by change of concentration of NaCl. It has been reported that the cause of the nervous symptoms is the depressant action of the Na on the central nervous system. The only gross anatomical change attributable to the experiments seemed to be the changes in the liver. Biochemical analysis of the blood for Na, K, and other substances might be of value in these animals.

TABLE 1. BLOOD SODIUM AND CHLORINE

Hog no.	Sample	Sodium	Chlorine
		mg. per 100 ml. serum	mg. per 100 ml. serum
476	a	Tube broken while centrifuging	
	b	314	317
472	a	336	349
	b	325	

Blood Sodium and Chlorine

In order to find out whether the blood of salt-poisoned pigs might contain abnormal amounts of sodium or chlorine, 50 cc. of blood in each of two samples were drawn from two of the affected pigs and analyzed for serum sodium and chlorine contents. Both pigs showed normal levels of the two salt constituents (table 1).

Summary and Conclusion

So far as we know, we here have a first recorded instance where it has been possible to salt-poison pigs experimentally, which effort in previous attempts had proved futile. It apparently could not be done by dry-feeding pigs or, for relatively few days and after salt starvation, by feeding simply wetted or soaked feed, followed by heavily salted feed, using abundant trough space all along.

Here were pigs that had been "conditioned" in the following manner:

1. The pigs were salt-starved for several months on rations that contained no animal protein concentrates and were inherently low in the elements of salt.

2. The pigs were fed swill which in itself prompts a rapid consumption of feed.
3. While trough space was sufficient when the pigs were small, the same length of trough brought on a crowded condition at feeding time when the pigs became larger.
4. Salt in amounts of 1.5 or 2.0 percent by weight of the customary swill was introduced all at once.
5. No additional drinking water at that time was available by means of which the pigs could have diluted the salt content of their digestive tract and of their body tissues. Vigorous scouring might have been a safety factor.
6. It required more than one such feeding or more than one day of excessive salt feeding before symptoms of poisoning became evident.

Where the above combination of circumstances, especially factors 1, 2, and 3, or even 2 and 3, happen to occur in combination, salt poisoning of pigs in connection with factor 4 may therefore be expected. A hog feeder should always be mindful of the danger of salt poisoning and should try in every way possible to avoid accidentally using excessive amounts of salt fed in a liquid form whether by way of whey, swill or brine. Salt should furthermore be fed consistently at all times in approved amounts or proportions when mixed in feed, unless the animals are given voluntary access to salt, preferably loose salt, which self-feeding has been found a good practice.

But also prominently in the picture is the necessity for adequate trough space at all times where the pigs are hand-fed swill.

Literature Cited

Grummer, R. H. and G. Bohstedt. 1947. Can "salt poisoning" kill pigs? Wis. Agr. Exp. Sta. Bul. 472.

Vestal, C. M. Apr. 15, 1947. Prevent salt hunger in hogs. Purdue Univ. Agr. Exp. Sta., Mimeo. No. 23, Dept. of Animal Husb.

THE RELATIONSHIP BETWEEN FEMININITY RATINGS AND REPRODUCTIVE PERFORMANCE IN GILTS

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FOR many years livestock breeders have given considerable attention to masculinity and femininity in the selection of animals to add to or to retain in their breeding herds and flocks on the contention that animals possessing these somewhat intangible qualities to the highest degree are the best breeders. Likewise, livestock judges have laid great stress upon strong development of masculinity and femininity on the premise that such qualities serve as a good partial index to the breeding worth of the animals.

A review of existing literature reveals many statements that express such views; however, there is apparently a complete absence of controlled data to substantiate or to refute such claims.

Since reproductive data were to become available on 79 gilts in three experiments designed to study the effects of certain nutritional and management factors on reproductive performance, it was decided to assign femininity ratings to these gilts and to study the correlation of these ratings with reproductive performance in an effort to secure controlled data on the relationship between femininity and reproductive performance.

Experimental Procedure

Femininity ratings were assigned to 79 gilts on three experiments. Experiment 1 included eight Poland China and twelve Hampshire gilts; experiment 2 included 40 gilts, of which 32 were purebred Hampshires and eight were crossbred Duroc-Hampshires; and experiment 3 included 19 Berkshire gilts.

In experiment 1, ratings were assigned first at weights of about 125 lb., and again at breeding time. Ratings assigned the second time were placed independently of the first set of ratings, and in only two cases did the ratings differ, two gilts being raised one rating at breeding. However, the ratings were assigned by the same man. Due to the consistency of the ratings in the first experiment, ratings thereafter were assigned only once, at weights between 125 and 150 lb.

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In experiment 1, ratings were broken down into six classifications, but this was reduced to only four in the following experiments on the assumption that most swine breeders can more easily recognize the breaks between four ratings than they can between six for a characteristic so difficult to describe as femininity. The four ratings used were: excellent (1), good (2), medium (3), and poor (4).

The femininity ratings were established from the appearance of the gilts from the shoulders forward, with greatest emphasis being placed directly upon the head. In order to qualify for the top rating, excellent, a gilt must exhibit extreme smoothness through the shoulders; her neck must be of moderate length with only sufficient thickness to blend smoothly into the shoulders; and the head must be broad and of medium length, extremely clean-cut and refined when viewed in profile, light and smooth in the region of the jowl, and wide and free from wrinkles in the region of the poll; it should possess refined ears of neat carriage and wide-open, clear, mild-appearing eyes. Varying degrees of deviation from these features result in the lower ratings of good, medium, and poor. No effort was made to include such items as length of body and teat development in the rating, for marked deviations were found to occur between the appearance of the head and such factors as these.

Gross correlations between femininity and number of normal embryos at 25 days after breeding, percentage of normal embryos at 25 days, and ovulation rate were secured for all three experimental groups. Since experiments 1 and 2 each represented four different feed-treatment-groups, the gross correlations were partitioned into between- and within-treatment-group correlations.

Results and Discussion

In the first experiment, four lots of five gilts each were used to test the effects of the various combinations of animal and vegetable protein and the presence and absence of an antibiotic (chloromycetin) on reproductive performance. Lots 1 and 2 received a ration of all vegetable origin, whereas lots 3 and 4 received a source of animal protein; lots 2 and 4 received the antibiotic, whereas lots 1 and 3 did not. Means and standard deviations for the ratings and various reproductive phenomena on the four treatment groups appear in table 1.

Data on the 20 gilts gave a within treatment group correlation of $-.774$ between femininity rating and number of normal embryos present at 25 days after breeding (table 2). This correlation was sig-

TABLE 1. MEANS AND STANDARD DEVIATIONS OF TREATMENT GROUPS FOR FEMININITY RATINGS AND REPRODUCTIVE PHENOMENA

Treatment Groups	No. Gilts	Femininity Rating		Number Normal Embryos		Percent Normal Embryos		No. Gilts	Femininity Rating		Ovulation Rate	
		Avg.	S.D.	Avg.	S.D.	Avg.	S.D.		Avg.	S.D.	Avg.	S.D.
Experiment I												
Lot 1	5	3.2	0.8	7.4	3.0	67.6	24.8	5	3.2	0.8	10.8	2.3
Lot 2	5	3.4	0.9	8.8	2.4	80.7	11.9	5	3.4	0.9	10.8	1.8
Lot 3	5	4.2	1.9	8.6	3.3	69.3	20.8	5	4.2	1.9	12.4	2.4
Lot 4	5	4.0	0.7	8.0	1.6	81.5	12.0	5	4.0	0.7	11.0	3.2
Experiment II												
Lot 1	5	1.8	0.8	10.2	1.9	90.4	9.6	10	2.1	0.9	10.6	1.2
Lot 2	5	3.0	0.7	7.2	1.8	71.1	18.7	10	2.9	0.9	9.4	1.3
Lot 3	5	2.8	0.8	6.2	3.5	54.8	30.9	10	2.5	0.7	10.3	1.9
Lot 4	5	2.6	0.5	8.0	2.0	81.3	13.6	10	2.9	0.6	9.5	1.4
Experiment III												
Early-spring	5	2.0	0.3	5.4	6.5	37.8	42.7	10	2.2	1.0	11.4	5.1
Late-spring								9	1.9	0.9	11.9	2.5
Total	45	3.0	1.4	7.8	3.2	70.5	25.8	79	2.7	1.1	10.7	2.6

nificant at the 1% level of probability, indicating that gilts with the strongest expression of femininity (as indicated by the lower rating numbers) tended to have a greater number of normal embryos present. Of additional interest is the fact that only one gilt was selected as being a typical number 1 (excellent) on femininity, and she topped the group of 20 gilts for number of normal embryos, with a total of 14. In this first experiment, there was only one gilt selected as representing a typical number 6 (poor); she was in the same treatment group as the above gilt but had only five normal embryos. The regression of number of normal embryos at 25 days on femininity rating indicates an increase of 1.7 embryos for each improvement of one unit in femininity rating (table 2).

The within-treatment-group correlation between femininity rating and percentage of normal embryos at 25 days was —.451, which did not prove significant when tested statistically. However, the within-treatment-group correlation of —.536 between femininity rating and ovulation rate was significant at the 5% level of probability.

In this first experiment, the effects of the significant correlation between femininity rating and ovulation rate and the high but nonsignificant correlation between femininity rating and percentage of normal embryos at 25 days resulted in a highly significant correlation between femininity rating and number of normal embryos at 25 days. It appears that higher ovulation rate in gilts with the strongest expression of femininity contributes more to the reproductive advantage of such gilts than does embryo survival.

In the second experiment, four lots of ten gilts each were used to test the effects of plane of nutrition and level of protein on reproductive performance. Lots 1 and 3 were full-fed on self-feeders, and lots 2 and 4 were hand-fed at sixty percent of the level of feed consumption for the full-fed lots. Lots 1 and 2 received a 20% protein ration, whereas lots 3 and 4 received a 14% ration. Means and standard deviations for the ratings and various reproductive phenomena in the four treatment groups appear in table 1.

Twenty of these gilts gave a significant gross correlation of —.596 between femininity rating and number of normal embryos. However, the within-treatment-group correlation dropped to —.407, which was not statistically significant (table 2). However, once again the gilt selected as the most outstanding on femininity topped the group on number of normal embryos at 25 days, with a total of 13. The regression coefficient for number of normal embryos at 25 days on femininity

TABLE 2. WITHIN-TREATMENT-GROUP CORRELATIONS AND REGRESSIONS OF REPRODUCTIVE PHENOMENA ON FEMININITY RATINGS

Experiment Number	No. Gilts	Number of Normal Embryos		Percent Normal Embryos		No. Gilts	Ovulation Rate	
		r	b	r	b		r	b
I	20	−.774**	−1.7	−.451	− 6.9	20	−.536*	−1.2
II	20	−.407	−1.3	−.234	− 6.3	40	−.419*	−0.8
III								
Early	5	−.878*	−5.8	−.848	−36.2	10	−.779**	−3.8
Late						9	−.812**	−2.2
Total gross r and b for all gilts	45	−.399**	−1.1	−.188	− 4.1	79	−.422**	−0.99

* Significant at 5% level.
** Significant at 1% level.
r is correlation coefficient.
b is regression coefficient.

rating indicates an increase of 1.3 embryos for each improvement of one unit in femininity rating.

Based on 20 gilts, a gross correlation of —.406 between femininity rating and percentage of normal embryos at 25 days dropped to —.234 for the within-treatment-group correlation. However, the highly significant gross correlation of —.496 between femininity rating and ovulation rate for 40 gilts was accompanied by a within-treatment-group correlation of —.419, which was significant at the 5% level of probability.

In no case in experiments 1 and 2 was the between-treatment-group correlation statistically significant.

In the third experiment, reproductive data were collected on a group of ten early-spring-farrowed and a group of nine late-spring-farrowed Berkshire gilts, which were part of a study of the effects of season of birth on reproductive performance in gilts.

A significant correlation of —.878 was found between femininity rating and number of normal embryos at 25 days for five early-spring-farrowed gilts. Two of these gilts received ratings of 1 and had the largest number of normal embryos at 25 days, with 12 and 13 embryos each. As a result of the wide variation in number of normal embryos in this lot, the regression coefficient indicates an increase of 5.8 embryos for each improvement of one unit in femininity rating.

The correlation between femininity rating and percentage of normal embryos, based on five early-spring-farrowed gilts, was —.848 which approached significance at the 5% level. Ten early-spring-farrowed gilts, however, gave a highly significant correlation of —.779 between femininity rating and ovulation rate. Thus there is quite apparent a trend for higher ovulation rates to contribute heavily toward an advantage in number of normal embryos in gilts of superior femininity.

Ovulation data for nine late-spring-farrowed Berkshires yielded a highly significant correlation of —.812 between femininity rating and ovulation rate. One gilt in this group that was selected as the most outstanding on femininity topped the group with 15 ova.

The gross correlation between femininity rating and number of normal embryos for a total of 45 gilts on the three experiments was —.399, which was significant at the 1% level of probability. The regression coefficient indicates an increase of 1.1 embryos for each improvement of one unit in femininity rating. For percentage of normal embryos, the gross correlation was —.188, which was not significant. However, for a total of 79 gilts on three experiments, the gross cor-

relation between femininity rating and ovulation was —.422, significant at the 1% level; and the regression coefficient of —0.99 indicates that an increase of one egg may be expected for each improvement of one unit in femininity rating.

Apparently, as judged by the number of normal embryos at 25 days after breeding, gilts with the strongest expression of femininity do have a tendency to produce larger litters. Such an increase in litter size may be due to an increased ovulation rate and/or to the attachment and development of a higher percentage of the fertilized ova in the gilts of excellent femininity.

As the development of the secondary female sexual characteristics, or femininity, is conditioned by estrogen, higher levels of secretion of this hormone would probably lead to a stronger expression of these characteristics. Fekete (1953) points out that the follicles and corpora lutea are the hormone-producing components of normal ovaries and concludes that the ovaries containing the largest quanity of these structures produce and supply the greatest hormonal stimulation. Robinson and Nalbandov (1951) report a highly significant correlation between the gonadotrophic potency of the pituitary gland and the follicular activity of the ovaries of gilts. Thus, increased pituitary activity apparently leads to increased follicular activity, which, in turn, results in the output of increased levels of estrogen, which could produce a stronger outward expression of femininity. It might be said, therefore, that femininity serves as a partial index to pituitary activity.

This hypothetical increase in follicular activity of a gilt of excellent femininity would result in the maturation of a greater number of ova, making a higher ovulation rate possible. The significant correlations between femininity ratings and ovulation rates reported in this study seem to indicate that this is true.

Although increased ovulation rates could account for the increased litter size in gilts of superior femininity, the additional factor of attachment and development of the embryo may be involved. Allen (1939) points out that the stimulation of the uterus by estrogen alone is not capable of supporting the life of an implanted embryo. In order to permit pregnancy, additional changes must be brought about by progesterone. Guthrie (1939) indicates that progesterone aids in the retention of the embryo if the ovum is fertilized, and this is in line with the findings of Glasgow *et al.* (1951) that with swine there is a significant correlation between the concentration of progesterone excretion products and the percentage of total ova shed resulting in

viable embryos. These workers also found a significant correlation between the percentage of total ova shed which were implanted and number of corpora lutea present. Apparently, with the presence of a greater number of corpora lutea, progesterone production is increased, and the increased levels of this hormone enhance the chances for implantation and development of the embryo. Thus, a gilt of excellent femininity should have a gerater number of corpora lutea formed as the result of a higher ovulation rate, and this might result in a larger percentage of the ova shed being represented by normal embryos.

The data secured in this study seem to indicate that the advantage in litter size in favor of gilts of excellent femininity is largely due to increased ovulation rates rather than to increased embryo survival. But high, though not significant, correlations between femininity ratings and percentage of normal embryos indicate that perhaps embryo survival does play a contributing role which might prove significant if tested with a larger number of gilts.

It is realized that the small number of ratings used to classify the varying degrees of femininity reduced the variability in this study. A more comprehensive classification could be used by experienced stockmen and livestock judges; however, due to the intangible qualities of the characteristic rated, a wider classification would be of doubtful value in the hands of many swine breeders.

All the ratings in this study were made by the research worker who was in close daily contact with the gilts. While this would correspond to the practice of a swine producer selecting gilts on femininity from within his own herd as brood sow replacements, it would be more desirable from the standpoint of further experimental proof to have the ratings in further experiments made independently by three qualified swine judges who have not been in close contact with the gilts and then use the average of their ratings. It may also be desirable to have such a rating committee place ratings on the gilts again after a 30-day lapse of time, to further check the repeatability of such ratings.

Summary and Conclusions

Femininity ratings, based entirely on appearance of the head and neck, were assigned to 79 gilts of the Poland China, Hampshire, and Berkshire breeds at weights of about 125 lb. to study the relationship between femininity ratings and reproductive performance.

Under the conditions of this study, it proved possible to select gilts

at weights of about 125 lb. for the largest number of normal embryos at 25 days after breeding, basing selection entirely on femininity.

The reproductive advantage obtained through selecting gilts on the basis of femininity is, apparently, reflected largely through increased ovulation rates rather than through decreased embryonic mortality.

The theory that femininity may serve as a rough index to pituitary activity is advanced and discussed.

Literature Cited

Allen, Edgar. 1939. Some investigations of reproduction in primates. Growth 3:221.

Fekete, E. 1953. A morphological study of the ovaries of virgin mice of eight inbred strains showing quantitative differences in their hormone producing components. Anat. Rec. 117:93.

Glasgow, B. R., D. T. Mayer and G. E. Dickerson. 1951. The colorimetric determination of the excretion products of progesterone and their correlation with certain phenomena of pregnancy in the sow. JOURNAL OF ANIMAL SCIENCE 10:1076 (abstract).

Guthrie, M. J. 1939. Reproductive cycles in animals with special reference to the bat. Growth 3:261.

Robinson, G. E., Jr. and A. V. Nalbandov. 1951. Changes in the hormone content of swine pituitaries during the estrual cycle. JOURNAL OF ANIMAL SCIENCE 10:469.

SOME EFFECTS OF SOURCE OF PROTEIN AND AN ANTIBIOTIC ON REPRODUCTIVE PERFORMANCE IN GILTS

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LITTER size in swine is largely dependent upon three factors: the number of ova released, the percentage of such ova fertilized, and the percentage of fertilized ova that develop to maturity. Therefore, any increase in ovulation and fertilization rates and decrease in embryonic mortality would result in an increase in number of pigs farrowed. Data presented by Squiers *et al.* (1950) and Robertson *et al.* (1951b) indicate that fertilization rates are quite high in swine, probably over 90 percent. Consequently, any noticeable increase in litter size will have to result from increased ovulation rates and decreased embryonic deaths. Accordingly, this experiment was initiated as part of a study designed to test the effects of various nutritional factors on reproductive performance. This phase of the study considered the effects of the various combinations of animal and vegetable protein and presence or absence of an antibiotic on ovulation rates and embryonic mortality.

Experimental Procedure

Twenty gilts, averaging three months of age, were divided into four lots of five pigs each—two Poland China and three Hampshire gilts to each lot.

The rations received by the various lots are shown in tables 1 and 2. Lots 1 and 2 received a vegetable protein ration, whereas Lots 3 and 4 received an added source of animal protein. Lots 1 and 3 received the basal rations unsupplemented, and Lots 2 and 4 received the basal rations supplemented with chloromycetin mycelial meal (0.5 percent). Rations for all lots were supplemented with vitamin B_{12} in order to eliminate its effect as a variable. All lots were self-fed by treatment groups in dry lot during the day and were grazed on Sudan pasture at night.

All gilts were checked daily for visible signs of onset of heat by examination of the vulva for increase in size and vascularity until they

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TABLE 1. RATIONS RECEIVED FROM START OF EXPERIMENT TO 125 POUNDS

Ingredients	Lot No. 1	2	3	4
	(parts)			
Ground milo	66.0	66.0	71.0	71.0
Supplement A [1]	30.0	30.0	....	....
Supplement B [2]	....	....	26.0	26.0
Vitamin B_{12} Supplement [3]	2.0	2.0	2.0	2.0
Ground limestone	1.5	1.5	0.5	0.5
Salt	0.5	0.5	0.5	0.5
Chloromycetin mycelial meal [4]	....	0.5	....	0.5

[1] soybean oil meal 80 lbs.; alfalfa leaf meal 20 lbs.
[2] meat scraps 40 lbs.; soybean oil meal 40 lbs.; alfalfa leaf meal 20 lbs.
[3] ground milo 96 lbs.; Dawe's B_{12} mix. 4 lbs.
[4] was substituted for an equivalent amount of mixed ingredient—contained 5.79 mg. B_{12} per gram and 4.2 mg. chloromycetin per gram—furnished by Parke Davis & Company.

were 120 days of age. After that time, they were checked twice daily, early morning and late evening, with a vasectomized board to determine the age of puberty. All gilts were allowed to come into their second estrus, at which time they were mated to a normal boar on the second day of heat. Each gilt was allowed only one service with the exception of one gilt, which was rebred six hours after an incomplete first service.

All gilts were slaughtered 25 days after breeding and data collected on ovulation rates for second heat, as determined by the number of corpora lutea present, and on embryo survival, including number, condition, and size of all embryos.

One gilt in Lot 2 died with enteritis early in the experiment, leaving most of the data based on 19 animals.

TABLE 2. RATIONS RECEIVED FROM 125 POUNDS UNTIL SLAUGHTER

Ingredients	Lot No. 1	2	3	4
	(parts)			
Ground milo	73.0	73.0	77.0	77.0
Supplement A	23.0	23.0	....	....
Supplement B	....	....	20.0	20.0
Vitamin B_{12} supplement	2.0	2.0	2.0	2.0
Ground limestone	1.5	1.5	0.5	0.5
Salt	0.5	0.5	0.5	0.5
Chloromycetin mycelial meal [1]	....	0.5	....	0.5

[1] was substituted for an equivalent amount of mixed ingredients.

Since chloromycetin (chloramphenicol) has in several instances been recorded as causing a fatal aplastic anemia in man, it was deemed desirable to check its effect on hemoglobin levels in swine. Consequently, blood samples were collected from each gilt at time of slaughter, and hemoglobin levels were determined by use of the colorimetric test for oxyhemoglobin.

An analysis of variance of all results was made, with adjustments for disproportionate subclass frequencies being made according to the method of Garber (1951).

Results and Discussion

Age of Puberty

Gilts in the animal protein lots attained puberty at an average age of 213.0 days as compared with 229.9 days for vegetable protein lots, for a difference of 16.9 days. This difference proved to be statistically significant at the 5% level of probability. This result is in agreement with data on poultry presented by Prentice *et al.* (1930) in which birds on a basal ration supplemented with vegetable protein did not lay their first egg until 186 days of age as compared with 137 days for birds receiving a source of animal protein.

Since work by Warnick *et al.* (1951), Robertson *et al.* (1951a), and Squiers and his co-workers (1952) indicates an increase in ovulation rate with an increase in sexual age in gilts, this earlier age of puberty for gilts receiving animal protein supplement could prove to be important where gilts are to be bred at about eight months of age. The animal protein gilts would have experienced an additional estrus and should ovulate a larger number of ova, thereby increasing the possibility of a larger litter.

Gilts receiving chloromycetin averaged 219.5 days at puberty as compared with 223.4 days for gilts not receiving the antibiotic. The difference of 3.9 days in favor of gilts receiving chloromycetin was not significant. Gilts receiving chloromycetin averaged 1.6 days older at puberty in animal protein lots and 9.3 days younger in vegetable protein lots than gilts not receiving the antibiotic. Although an interaction between source of protein and chloromycetin is indicated, the interaction was not statistically significant.

Ovulation Rates

The average second heat ovulation rate of all animals was 11.3 ova, with a range of 8 to 16 and a standard deviation of 2.4.

Gilts receiving animal protein averaged 11.7 ova at second estrus as compared with 10.8 ova for gilts on vegetable protein. The difference of 0.9 ova, although suggesting an advantage in favor of animal protein, was not statistically significant.

Gilts receiving chloromycetin averaged 10.9 ova at second estrus in comparison to 11.6 ova for gilts not receiving the antibiotic, giving a difference of 0.7 ova.

Number of Normal Embryos

Seventeen gilts provided data on the number of embryos at 25 days after breeding. (Two gilts in Lot 3 remained in heat for approximately 48 hours after breeding and failed to settle. Although gilts occasionally will stay in heat three days or longer, these gilts were not rebred on the third day—in keeping with general practice in such cases—due to the decision at the start of the experiment to allow only one service per gilt.) The average number of normal embryos for all gilts was 8.2, with a range of 3 to 14 and a standard deviation of 2.7. Normality of embryos was based on the presence of a heart beat or on normal size and freedom from apparent degenerative changes in the absence of an observed heart beat.

Gilts receiving vegetable protein averaged 8.0 normal embryos in contrast to 8.5 for gilts on animal protein, and gilts receiving chloromycetin averaged 8.6 as compared to 8.0 for gilts not receiving the antibiotic. In no case, however, was the difference significant. As table 3 indicates, the gilts on vegetable protein without the antibiotic supplement averaged only 7.4 normal embryos at 25 days after breeding whereas all other lots averaged over eight.

Percent of Normal Embryos

For the seventeen gilts, 74.7 percent of the ova shed were represented by normal embryos, with a standard deviation of 19.3 percent.

Gilts on vegetable protein averaged 74.1 percent normal embryos as compared with 75.4 percent for gilts receiving animal protein. Gilts receiving chloromycetin, however, averaged 81.0 percent as compared with only 68.5 percent for those gilts not receiving the antibiotic. Although this advantage of 12.5 percent in favor of gilts receiving chloromycetin did not prove to be statistically significant, it may be suggestive of a favorable effect of the antibiotic on embryo survival. Carpenter (1951) found that aureomycin did not affect litter size, and recently De Pape and co-workers (1953) found that aureomycin made no significant improvement in number of pigs farrowed. Bell (1952)

found no improvement in number of pigs farrowed upon the addition of an APF supplement but did find a slight trend toward lower percentages of stillborn pigs in favor of the APF supplement.

Hemoglobin Levels

According to Hart and co-workers (1929), the blood of a mature sow has a hemoglobin content of 8 to 11 gm. per 100 cc.

TABLE 3. SUMMARY OF EFFECTS OF SOURCE OF PROTEIN AND CHLOROMYCETIN

		Phenomena				
				Normal Embryos at 25 days		
Lot	Treatment	Age at Puberty	Ovulation Rate 2nd Heat	Percent	Number	Hemoglobin gm./100 cc.
I	Veg. prot.-no chloromycetin	234.6	10.8	67.6	7.4	10.86
II	Veg. prot.-chloromycetin	225.3	10.8	80.5	8.8	11.20
III	An. prot.-no chloromycetin	212.2	12.4	69.3	8.6	10.88
IV	An. prot.-chloromycetin	213.8	11.0	81.5	8.3	11.64
	Avg. for Veg. prot.	229.9	10.8	74.1	8.0	11.03
	Avg. for An. prot.	213.0	11.7	75.4	8.5	11.26
	Difference	16.9*	0.9	1.3	0.5	0.23
	Avg. for no chloromycetin	223.4	11.6	68.5	8.0	10.87
	Avg. for chloromycetin	219.5	10.9	81.0	8.6	11.42
	Difference	3.9	0.7	12.5	0.6	0.55

* Significant at the 5% level of probability.

In this experiment, hemoglobin levels for gilts receiving vegetable and animal protein in combination with chloromycetin were 11.20 and 11.64 gm. per 100 cc. of blood, respectively, compared with 10.86 and 10.88 for gilts on vegetable and animal protein without the antibiotic. Statistically, the difference is not significant, and it is apparent that there was no detrimental effect of chloromycetin on hemoglobin levels. Actually, gilts receiving chloromycetin averaged slightly higher hemoglobin levels than gilts not receiving the antibiotic. This is in keeping with results obtained with another antibiotic, aureomycin, by several other research workers (Burnside *et al.*, 1951; and Edwards *et al.*, 1950). Thus, apparently chloromycetin mycelial meal does not have the toxic effect on swine that the purified antibiotic has exhibited in some cases of aplastic anemia in humans.

Summary and Conclusions

Four lots of five gilts each were used to test the effects of the various combinations of animal and vegetable protein and the presence or absence of chloromycetin on reproductive performance in gilts.

In the case of source of protein, the only significant difference demonstrated was a stimulating effect toward an earlier age of puberty in favor of animal protein. Although not statistically significant, an increase in ovulation rate in favor of animal protein was indicated.

No significant effects of chloromycetin on reproductive performance were demonstrated. However, the greater percentage of ova shed represented by normal embryos in lots receiving chloromycetin suggests the antibiotic may exert a favorable influence on embryo survival.

There was no detrimental effect of chloromycetin on hemoglobin levels.

Literature Cited

Bell, J. M. 1952. Animal protein factor in swine rations. Scientific Agr. 32:135.

Burnside, J. E., R. H. Grummer, P. H. Phillips, and G. Bohstedt. 1951. The effect of vitamin B_{12} and aureomycin when fed in high, intermediate, and low protein levels to swine. JOURNAL OF ANIMAL SCIENCE 10:1042 (abstract).

Carpenter, L. E. 1951. Effect of APF concentrate containing aureomycin on gestating, lactating, and growing swine. JOURNAL OF ANIMAL SCIENCE 10:657.

De Pape, J. G., W. H. Burkitt, and A. E. Flower. 1953. Dehydrated alfalfa and antibiotic supplements in gestation-lactation rations for swine. JOURNAL OF ANIMAL SCIENCE 12:77.

Edwards, H. M., T. J. Cunha, G. B. Meadows, R. F. Sewell, and C. B. Shawver. 1950. Observations on aureomycin and APF for the pig. Proc. Soc. Exper. Biol. & Med. 75:445.

Garber, M. J. 1951. The influence of sire, dam, and hatching date on growth of single comb White Leghorn pullets from hatching to eighteen weeks of age. Ph.D. thesis. Agricultural and Mechanical College of Texas.

Hart, E. B., C. A. Elvehjem, H. Steenbock, G. Bohstedt, and J. M. Fargo. 1929. Anemia in suckling pigs. Wis. Agr. Expt. Sta. Bul. 409.

Prentice, J. H., R. G. Baskett, and G. S. Robertson. 1930. The nutrition of the chick and its effect on growth, maturity, egg production, and mortality. IV World's Poultry Congress. London. No. 37, 224.

Robertson, G. L., R. H. Grummer, L. E. Casida, and A. B. Chapman. 1951a. Age of puberty and related phenomena in outbred Chester White and Poland China gilts. JOURNAL OF ANIMAL SCIENCE 10:647.

Robertson, G. L., L. E. Casida, R. H. Grummer, and A. B. Chapman. 1951b. Some feeding and management factors affecting age of puberty and related phenomena in Chester White and Poland China gilts. JOURNAL OF ANIMAL SCIENCE 10:841.

Squiers, C. D., G. E. Dickerson, and D. T. Mayer. 1950. Factors affecting litter size during early pregnancy in certain strains of swine and their crosses. JOURNAL OF ANIMAL SCIENCE 9:683.

Squiers, C. D., G. E. Dickerson, and D. T. Mayer. 1952. Influence of inbreeding, age and growth rate of sows on sexual maturity, rate of ovulation, fertilization and embryonic survival. Mo. Agr. Exp. Sta. Res. Bul. 494.

Warnick, A. C., E. L. Wiggins, L. E. Casida, R. H. Grummer, and A. B. Chapman. 1951. Variation in puberty phenomena in inbred gilts. JOURNAL OF ANIMAL SCIENCE 10:479.

ABSTRACTS OF PAPERS TO BE PRESENTED AT THE FORTY-SIXTH ANNUAL MEETING OF THE AMERICAN SOCIETY OF ANIMAL PRODUCTION

The meetings are scheduled to be held at the Hotel Sherman in Chicago on November 26 and 27, 1954. Abstracts have been submitted for the following papers.

Breeding and Genetics Section

Charles R. Henderson, *Chairman*

INFLUENCE OF SEASON OF BIRTH ON GROWTH RATE AND SURVIVAL OF PIGS. Arthur D. Allen and John F. Lasley, *University of Missouri.*

The influence of the season of farrowing on the performance of 172 sows and 1,635 pigs was studied at the Missouri Station. An average of 1.32 more pigs per litter ($P < .01$) was farrowed in the fall than in the spring, but the difference decreased to 0.86 pigs per litter at 56 days ($P < .01$) and to 0.04 pigs at 154 days. Pigs farrowed in the fall averaged 0.20 lb. heavier at birth ($P < .01$), but they were no heavier at 56 and 154 days of age than spring-farrowed pigs. The time of farrowing in the spring greatly influenced pig survival, mostly because of extreme temperature changes, but did not have such a great influence on growth rate. The time of farrowing in the fall had a definite influence on the performance of the pigs. Survival and growth rate both decreased as the farrowing season progressed. Between August 10 and September 30, there was an average decline of 0.34 lb. per pig in weight at weaning and 1.41 lb. per pig in weight at 154 days of age for each day later in the season that the pigs were farrowed.

COMPARISONS OF TOPCROSSES BY INBRED AND LINECROSS BOARS WITH NONTOPCROSSES IN TWO-SIRE FARM HERDS. G. E. Bradford, A. B. Chapman and R. H. Grummer, *University of Wisconsin.*

Inbred and linecross boars developed at Wisconsin and other experiment stations cooperating in the Regional Swine Breeding Laboratory have been compared with outbred boars in two-sire farm herds. Only comparisons in which both kinds of matings within a herd were straightbred or both were crossbred have been used. Earlier findings in this experiment, that topcrosses by inbred boars do not differ significantly on the average from nontopcrosses in viability and rate of gain, but do show a significant improvement in gilt productivity, have been substantiated by additional inbred boar comparisons. Topcrosses by linecross boars show an advantage over nontopcrosses in viability and rate of gain, but the gilts by linecross boars show no over-all advantage in productivity. Comparisons within herds in which different inbred or linecross boars were tested in consecutive years indicate that litters out of gilts sired by inbred boars show approximately the same advantage whether these litters are sired by inbred, by linecross, or by outbred boars.

INTERRELATIONS OF MILK PRODUCTION AND BREEDING EFFICIENCY IN DAIRY COWS. Grant M. Carman, *Iowa State College.*

The primary purposes of this study were (1) to obtain unbiased estimates of the repeatability and heritability of three measures of breeding efficiency: time to first estrus following parturition; time from first breeding to conception, number of services to conception, and (2) to study the relationship between breeding efficiency and production level. Data from 1,646 lactation intervals of 763 cows in two herds were used, all cows having completed at least one lactation and being successfully bred during that lactation. The means of the three measures were 55.4 ± 1.1, 28.0 ± 1.07, and 1.8 ± 0.04 in herd 1, 71.0 ± 1.8, 42.0 ± 2.7, and 1.7 ± 0.04 in herd 2. Cows freshening in March took longer to come into heat (65 days in herd 1 and 83 days in herd 2) than in any other month, while those freshening in September came into heat more quickly (45 days in herd 1 and 57 days in herd 2). Only the period from parturition to first estrus was found to be correlated significantly with preceding production. Repeatabilities of days to first estrus were 0.15 and 0.27; for days to conception 0.08 and 0.02; for services to conception 0.06 and 0.06 in herds 1 and 2, respectively. Heritabilities of the same criteria were $-.06 \pm .11$ and $-.03 \pm .18$; $.08 \pm .08$ and $-.09 \pm .11$; $.08 \pm .09$ and $-.15 \pm .09$ in herds 1 and 2. This investigation indicates that the repeatability and heritability of breeding efficiency, when measured by the three variables used, are zero, or nearly so. More progress could be made in selecting for other traits of economic importance for which repeatability and heritability values are higher.

ENVIRONMENTAL FACTORS AFFECTING BODY DIMENSIONS IN YEARLING HAMPSHIRE SHEEP. D. W. Cassard, P. W. Gregory, W. C. Weir and J. F. Wilson, *University of California.*

In mutton breeds of sheep much attention is given in selection to body size and shape. Because environmental causes of variation in these characters obscure heritable differences estimates are needed of these effects. A study of body measurements taken on 227 Hampshire yearling ewes at the University of California at Davis over the past 17 years was undertaken to estimate the effects at this stage of differences in age and inbreeding, of being a twin rather than a single, and of having a two-year-old dam rather than a mature one. Average age was 440 days; inbreeding coefficients ranged from 0 to 37.5% with an average of 3.4%. Constants fitted by least squares procedure indicate that for each day difference in age, older animals were larger than younger ones by .21 lb. weight, .06 cm. in heart girth, .04 cm. in round measurement, .01 cm. in withers height, .02 cm. in chest depth and chest width and .01 cm. in loin width. For each 1% higher inbreeding coefficient, animals were smaller by .58 lb. body weight, .20 cm. in heart girth, but less than .10 cm. in other measurements. Twins were smaller than singles by 8.22 lb. in weight, by 1.69 cm. in heart girth, by 1.42 cm. in round measurement, 1.10 cm. in withers height, and less than 1 cm. in other measurements. Differences due to age of dam were small, the largest being .51 cm. in heart girth in favor of offspring of older dams.

THE INHERITANCE OF DWARFISM IN A COMPREST HEREFORD HERD. Doyle Chambers, J. A. Whatley, Jr. and D. F. Stephens, *Oklahoma Agricultural Experiment Station.*

In 1949 a study was initiated to compare the reproductive performance of large- and small-type Hereford cattle. Twenty-five weanling heifer calves of each type were selected and moved to the Fort Reno Station. The small-type heifers and

bulls used were of "Comprest" breeding. Three "Comprest" bulls were bred to the cows of the "Comprest" line for the 1952 calf crop and six dwarf calves were dropped. Each bull sired one or more dwarfs. Two of the same bulls were bred to this herd for the 1953 calf crop and five dwarf calves were produced. Again both of the bulls sired dwarf calves. From 45 such matings, thirty-seven calves were dropped, eleven of which were definitely dwarfs. Three of these dwarf calves were of the "crooked-legged" type; five of them were "straight-legged," and three dwarfs were very extreme but were not further classified because they were still-born. Four early stage abortions or resorptions were known to have occurred. In 1953 two known dwarf-carrier Hereford bulls of noncomprest breeding and one known dwarf-carrier Angus bull were bred to 24 of these Comprest cows and seven yearling heifers produced in this line. Twenty-seven of them calved; six of the calves were dwarfs, none of which were crooked-legged. One resorption or abortion occurred during early pregnancy. Five of the dwarfs were Herefords, and one was a crossbred Angus X Hereford. These results indicate either that the genes responsible for dwarfism in "compress" and conventional Hereford and Angus cattle may be allelic or that the "Comprest" cattle in this test also carried in high frequency the recessive dwarf gene found in non-comprest cattle.

LIVE HOG PROBES AT VARIOUS SITES, WEIGHTS AND AGES AS INDICATORS OF CARCASS MERIT. J. G. De Pape and J. A. Whatley, Jr., *Oklahoma Agricultural Experiment Station.*

Probe backfat thickness measurements were made behind the shoulder and over the loin on one side of 111 pigs and behind the shoulder, over the loin and over the back on both sides of 72 pigs at a slaughter weight of about 210 lb. In addition, 12 Duroc, 11 Beltsville No. 1 and 24 pigs of the reciprocal crosses between these breeds were probed at the three sites, but on one side, at monthly intervals from weaning at 56 days to market weight.

Intra-breed and sex correlations with 104 degrees of freedom of the average of probes behind the shoulder and over the loin with carcass backfat thickness at the seventh rib was .36. In the second group the average of eight carcass backfat measurements was correlated .69 with the average of six probes. The average of three probes on one side at 140 days of age was correlated .51 with carcass backfat thickness. Probes taken at ages younger than 140 days had very low predictive values.

Intra-breed and sex correlations with 68 degrees of freedom of average carcass backfat thickness and average of six probes at slaughter weight with the following carcass traits were: percent lean cuts — .66, — .57; percent primal cuts — .58, — .67; loin lean area — .28, — .26; ham specific gravity — .30, — .36; carcass index — .43, — .55; and carcass length — .11, and — .33 respectively.

Measurable differences between breeds in backfat deposition at 140 days and older were observed. A combination of probes behind the shoulder and over the loin on both sides was found to be the most useful of those studied.

AN ANALYSIS OF WRY CALVES IN CALIFORNIA BEEF HERDS. P. W. Gregory, *University of California.*

During recent years a high percentage of "wry" calves have occurred in several areas in California resulting in serious financial loss. The incidence was especially high in certain ranges of Butte and Lassen Counties. Reports indicate that similar anomalies may be occurring in other western states. At least two distinct morphological wry types were observed. The first type is rather uniform in appearance and occurs in animals of both Hereford and Angus breeding. This type possesses

distinct dwarf features but all afflicted animals are characterized by crooked forelegs. The second type does not possess dwarf characteristics and has only been observed in animals of Hereford breeding. The gross manifestations are a crooked spine and crooked forelegs with enlarged knee joints. Other features that may be present are wry neck and cleft palate. There is considerable variation in the expression of each of these characteristics. The incidence of this second wry type may exceed 15% in some herds. The evidence indicates that the type that possesses dwarf characteristics combined with crooked forelegs results from a specific interaction of one comprest gene with two conventional dwarf genes. The possible factor interactions of the alleles occupying the comprest and dwarf loci are discussed. There is no evidence that indicates the second wry type is conditioned by heredity. In fact, the available data suggests that the anomaly may be attributable to faulty nutrition at a critical period during gestation.

THE RELATIONSHIP BETWEEN BODY WEIGHT, DAILY GAIN, AND EFFICIENCY OF FEED UTILIZATION IN BEEF CATTLE. J. E. Grizzle and C. M. Kincaid, *Virginia Polytechnic Institute.*

A study of the relationship between weight and daily feed consumption from data collected on 121 bulls and 66 steers individually fed over a five-year period showed that the linear regression of daily feed consumption on weight explained a highly significant amount of the variation in feed consumption. After adjustment for linear regression, the data showed highly significant differences among periods and animals within groups. Group differences were highly significant in the 1950–51 and 1951–52 bulls and steers, and the group-period interactions were significant in all animals fed except the 1947–48 bulls and 1951–52 steers. Correlations between rate of gain and efficiency based on (1) observed feed consumption and (2) feed consumption adjusted for regression on weight ranged: for (1) from —0.05 to 0.63, and for (2) from 0.77 to 0.88. It appears from the data secured in this experiment that rate of gain is a better measure of efficiency than the ratio of gain to feed consumption unless adjustment is made for differences in weight or constant end weights are used. Pounds of feed per pound of gain as a measure of efficiency in feeding trials may be misleading. In general, the younger and therefore smaller animals tended to be more efficient because they ate less feed than larger ones. In these trials, rate of gain tended to be more or less constant for the period from weaning to fifteen months of age.

EFFECT OF THAWING TEMPERATURE AND EXTENDER COMPOSITION ON THE FERTILITY OF FROZEN BULL SEMEN. Harold D. Hafs and F. I. Elliott, *American Foundation for the Study of Genetics.*

Motility studies suggested that thawing at 40° C. may be superior to 5° C., and intermediate temperatures inferior to both. A field trial with semen from four Holstein bulls involving nine technicians and 661 first services gave the following results: 5° C.: 63.0%; 20° C.: 61.4%; and 40° C.: 66.5% sixty to ninety day nonreturns. The differences are not significant statistically. The Polge method includes the addition of glycerol in citrate buffer to the egg yolk-citrate-semen mixture. Motility studies indicated an improvement in post-freeze motilities where the glycerol was added in an egg yolk-citrate medium. A field trial using semen from seven Holstein bulls and involving eight technicians and 1,563 first services resulted in 28–35 day nonreturn rates of 73 and 78% for the control and revised diluters respectively. The difference is significant at the 5% level of probability.

THE EFFECT OF INBREEDING ON GENETIC COMPONENTS OF GENERAL AND SPECIFIC COMBINING ABILITY. C. R. Henderson, *Cornell University.*

It has been shown by Cockerham that the total genetic variance of a population can be partitioned into parts described as additive, dominance, additive × additive, dominance × dominance, additive × additive × dominance, etc. It is the purpose of this report to describe the effect of inbreeding on the contribution of these different components of genetic variation to general and specific combining ability variance in crosses among inbred lines. Let σ^2_{ij} be the variance in an outbred population of a component due to the interaction of i alleles each at a different loci with j pairs of alleles. For example, σ^2_{10} denotes additive, σ^2_{01} dominance, and σ^2_{12} additive × dominance × dominance. Then in crosses among unselected lines with inbreeding coefficient F, the general combining ability variance is the sum of all $(\frac{F}{2})^i\sigma^2_{i0}$. The specific combining ability variance is the sum of all $(1-\frac{1}{2^{i-1}})F^i\sigma^2_{i0}$ and of all $F^{i+2j}\sigma^2_{ij}$, where j does not equal 0. It is apparent that higher order gene interactions contribute little to specific combining ability and consequently to the success of the "hybrid corn" method in animal breeding unless F is near 1.

USE OF THE CONTEMPORARY HERD AVERAGE IN APPRAISING PROGENY TESTS OF DAIRY BULLS. C. R. Henderson, H. W. Carter and J. T. Godfrey, *Cornell University.*

It had been found in previous studies by the authors that approximately one-half of the variance for milk and butterfat production of artificially sired cows can be attributed to differences among herds. Consequently it appeared that artificial insemination progeny tests based on small numbers of daughters would contain at best a large random error due to herd differences. Further, if any selection of bulls for particular kinds of herds was practiced, an important bias would result. An appropriate correction of each record for the herd in which it appears would reduce both sampling errors and bias. First lactation records of 7,850 artificially sired daughters of 128 Holstein bulls were used to develop such a correction factor. The intra-sire regression of daughter record on the contemporary herd average (excluding the daughter in question) was estimated. All records were converted to a 305 day, m. e., 2 × basis. The esimate of this regression was approximately .6. To adjust the progeny average in accordance with this estimate of the herd effect, the average production of the progeny of a particular bull is reduced .6 of the amount by which the unweighted mean of the contemporary herd averages exceeds the average of all herds in the population.

INDIVIDUAL VERSUS GROUP FEEDING OF STEERS ON FULL FEED. C. M. Kincaid, J. S. Copenhaver and F. S. McClaugherty, *Virginia Polytechnic Institute.*

Individual feeding was compared with group feeding of steers within sire progenies over a 112-day feeding period. From each of eight sire progenies, three steers were randomized to individual stalls and the remainder to groups with a total of 24 fed individually and 39 fed in two groups. From weaning in October to the start of this test, all animals had been on full feed with access to a surplus of feed at all times. The ration was the same for all experimental animals at all

times during this test and was a mixture of ground hay, ground ear corn, and oil meal. All experimental animals were confined in the barn with access to feed throughout each day, except for two exercise and watering periods of approximately an hour each. The respective averages for individual and group feeding were: daily gain *1.87* and *1.89* lb.; slaughter grade 11.51 (low choice) and 11.80 (low choice); and daily feed intake *18.22* and *18.03* lb. The differences in daily gain; slaughter grade, and daily feed intake were 0.02 ± 0.06 lb., 0.092 ± 0.053 grades and 0.19 ± 0.49 lb., respectively. The data indicate that *ad. lib.* feeding of a ground mixed ration gives essentially the same results when steers are fed in individual stalls as when fed in groups.

ASSOCIATION BETWEEN INBREEDING AND ANTIGEN OCCURRENCE IN INBRED LINES OF HEREFORD CATTLE. N. S. Kushwaha, L. C. Ferguson and H. H. Stonaker, *Colorado A. & M. College.*

This study was undertaken to find whether or not there is an increase in homozygosity of genes determining the presence of bovine blood antigen with increased inbreeding. The study was based on the blood types of 343 animals in 11 inbred lines of Hereford cattle. The average inbreeding and the blood antigen number for all the animals was found to be .24 and 8.8 respectively. Each animal was tested for the presence or absence of 28 antigen. The average inbreeding of those animals tested in 1950, 1951, 1952, and 1953, was .20, .23, .25, and .29 and corresponding mean number of blood antigen found was 8.8, 9.3, 9.2, and 7.9.

A general regression of blood antigen number on inbreeding percentage of $-.014 \pm .0017$ was found. Variances due to sires and years in which the blood typing was done were held constant. This regression indicated that within these lines an increase of 10% inbreeding could be expected to result in a loss of about 1.4 blood antigen.

Thus the study indicated that the slope of the regression was in agreement with that expected. Increased inbreeding would be expected to decrease the heterozygous class and increase both homozygous classes. This would have been expected to bring about an average decrease in the frequency of occurrence of the phenotypic expression of the various antigens.

PERFORMANCE OF SELECTED RAMS IN RELATION TO THAT OF THEIR UNSELECTED HALF-SIBS. W. T. Magee, *Texas Agricultural Experiment Station.*

Data for this study came from 22 pens of paternal half-sibs (4 selected rams per pen) bred and tested on a high plane of nutrition at Substation No. 14, Sonora, Texas, during the last five years and all their half-sisters carried on pasture, an average of 13 per sire group. The regression of the clean fleece weight of the ewe lambs on their half-brothers was .15, .14, .06, .27, .09 and .11 for the years 1950 through 1954 and average of all years, respectively. The corresponding regressions for grease fleece weight were .12, .31, — .01, .16, .11 and .14. For staple length, the regressions were 2.12, .41, .36, .48, .52 and .44 (excluding the first year's regression from the average). The regressions of the weaning weight of all lambs (rams and ewes) by a ram on the gain of his selected sons were .12, .38, — .01, — .17, — .12 and .04. The expected regression, using the method of path coefficients, of the clean or grease fleece weight of the ewes on a random sample of their half-brothers was .10. The expected regressions for staple length and growth rate were .36 and .25, respectively. The negative regression coefficient for 3 of the 5 years for growth rate indicates that gain on test is not closely related to weaning weight. The observed regression coefficients equalled or exceeded the expected ones for fleece

weights and staple length, thus indicating that selecting the rams that went on test did not destroy the accuracy of the test as a progeny test for their sires.

SOME ENDOCRINE ASPECTS OF DWARFISM IN BEEF CATTLE. Thomas J. Marlowe and Doyle Chambers, *Oklahoma Agricultural and Mechanical College.*

Studies were conducted to determine the thyrotropic, gonadotropic, adenocorticotropic and growth hormone content of the pituitaries from dwarf and normal calves of the Hereford and Angus breeds. Injection of glandular tissue suspensions from 25 dwarf and 23 normal calves into 410 baby chicks failed to reveal significant differences in the thyrotropic hormone potency of dwarf and normal calf pituitaries. Pituitary suspensions from 6 dwarf and 6 normal calves injected into 42 hypophysectomized immature female rats gave a highly significant difference in weight of uteri in favor of the dwarf calf glands. Injections of pituitary suspension from 26 dwarf and 17 normal calves into 54 "plateaued" female rats produced an increase in body weights of 16.6 and 11.3 grams, respectively. This difference was significant. Two assays in which 92 hypophysectomized immature female rats were injected with pituitary suspensions from 21 dwarf and 16 normal calves also strongly indicated a greater potency of growth hormone in the pituitaries of dwarf calves. The measures of responses included (1) increase in body weight (2) increase in tail length and (3) increase in width of the epiphyseal cartilage of the tibia. The increase in body weights during a 14-day injection period was 18.0 and 9.7 gm. for dwarf and normal calf pituitaries, respectively. This difference was highly significant. Assays for ACTH potency of 25 dwarf and 21 normal calf pituitaries using both baby chicks and hypophysectomized immature female rats failed to reveal any significant differences between them in adrenal weights. In each case the control roup was significantly lower than the injected groups.

EFFECTS OF SEASON OF BIRTH, SEX, BIRTH WEIGHT AND DIET ON GROWTH OF DAIRY CALVES. T. G. Martin, N. L. Jacobson and P. G. Homeyer, *Iowa State College.*

Data on 593 calves from the Iowa State College dairy herd were classified according to breed, season of birth, sex and ration during initial eight weeks (five broad classifications based on estimated growth promoting ability). Within classification coefficients of correlation of birth weight with eight-week and one-year weight gains were 0.159 ± 0.046 and 0.319 ± 0.067, respectively. Least squares analysis of eight week gains indicated that breed, sex, and ration effects were highly significant ($P < 0.01$) whereas season of birth had no significant effect on gain. Correcting eight-week gains for variation in birth weight markedly reduced sex and breed effects but both remained significant, indicating definite breed and sex differences independent of variation in birth weight. Least squares analysis of one-year gains was based only on Holsteins; therefore, breed differences could not be estimated. Sex and ration effects were found to be larger than effects of season of birth, though all were significant ($P < 0.05$). Correction of one-year weight gains for variation in birth weight had little effect on sex differences and essentially no effect on season and ration differences.

LENGTH OF GESTATION IN HEREFORD COWS AND ITS RELATION TO PERFORMANCE. F. J. Rice, A. M. Kelley and J. F. Lasley, *University of Missouri.*

The length of gestation and its relationship to birth and weaning weight of calves was studied in 443 Hereford range cows bred to 8 different bulls by artificial

insemination during a single season. The mean length of gestation for 212 heifer calves was 286.5 ± 6.18 days and for 231 bull calves was 287.3 ± 6.21 days but this difference was not significant. Cows bred to different bulls varied in gestation length from 284.0 ± 5.85 to 289.6 ± 4.91 days ($P < .01$). The mean birth weight of 132 heifer calves was 70.71 ± 7.88 lb. and for 191 bull calves was 73.0 ± 8.10 lb. ($P < .01$). Birth weights of calves sired by 8 different bulls varied from 70.0 ± 8.86 to 78.3 ± 5.79 lb. ($P < .01$). As the length of the gestation period increased, the birth weight of the calves also increased ($P < .01$). All weaning weights were corrected to a 205 day basis, and the mean for 323 calves was 330.1 ± 46.1 lb. Bull calves weighed an average of 28.8 lb. heavier than heifer calves ($P < .01$). The sire also had a significant effect ($P < .01$) on the weaning weight of the calves with the extreme difference between sire groups being 42.8 lb.

HERITABILITY OF SOME PRE- AND POST-SLAUGHTER CHARACTERISTICS IN BEEF CATTLE. C. E. Shelby, R. T. Clark, J. R. Quesenberry and R. R. Woodward, *U. S. Range Livestock Experiment Station.*

Record of Performance procedures have stressed various growth and carcass characteristics thought to be of economic importance. In order to evaluate their usefulness as criteria for selection, the genetic and environmental relationships existing between and within these characteristics should be known. Intra-year and-line heritabilities for a number of these characteristics were computed by the method of paternal half-sib correlation. They are based on data collected during ten years (1941–1952) of R. O. P. testing at the U. S. Range Livestock Experiment Station at Miles City, Montana. The data consist of the records of 635 steers from grade cows mated to 88 sires from nine lines. The effects of differences betweeen years, between lines within years, and between sires within lines were separated by the analysis of variance, while the effects of age at weaning, and age and final weight at the completion of the feedlot period were removed by the analysis of covariance. The following estimates of heritability were obtained: birth weight 71.6, weaning weight 22.8, gain in the feedlot 60.4, final weight at the end of the feedlot period 84.0, efficiency of feed utilization 21.8, slaughter grade 42.1, shrink 91.4, dressing percentage 72.7, carcass grade 16.4, color of eye 31.2, area of eye 71.5, and thickness of fat 37.7.

A PRELIMINARY REPORT OF CROSSING SEVEN INBRED LINES AND FOUR NON-INBRED BREEDS OF SWINE. G. W. Sherritt, H. O. Hetzer, E. Cobb and J. L. Gobble, *The Pennsylvania State University.*

Gilts of four breeds—Berkshire, Chester White, Hampshire, and Poland—were crossed in all possible combinations with the boars of seven inbred lines. The inbred lines were Landrace (L), Landrace-Chester White (L-CW), Landrace-Duroc (L-D), Landrace-Duroc-Hampshire (L-D-H), Landrace-Duroc-Hampshire-Yorkshire (L-D-H-Y), Landrace-Large Black (L-LB), and the Landrace-Poland China (L-PC) lines developed at Beltsville. The work was conducted on the farms of purebred breeders in Pennsylvania and equal numbers of purbred litters were used for comparison on each farm. The first cross was duplicated in the first and second years. The second phase of the experiment, the third and fourth years, was a comparison of crossbred gilts (American breed X Beltsville inbred line) and their progeny with purebred gilts and their progeny. One-half of the crossbred gilts were mated to a boar of the parent American breed and one-half to a Montana No. 1 boar on each farm. The first-cross pigs and the pigs produced by the crossbred gilts of both groups were heavier at weaning and 140 days of age, and gained

at a faster rate than the purebred pigs. The crossbred gilts of both groups produced larger litters at farrowing and weaning.

OBSERVATIONS ON REPRODUCTION, GROWTH, FEED UTILIZATION AND GRADES OF INBRED AND OUTBRED HEREFORD CATTLE, H. H. Stonaker, *Colorado A. & M. College.*

Reproduction, growth, feed utilization and grades were compared for inbred and outbred Hereford cattle. Records were on the contemporary inbred and outbred progeny born in 1951, 1952, and 1953 by herd sires within 12 inbred lines. There were 284 matings within 12 inbred lines and concurrently 275 matings of these same sires with unrelated, outbred females. Ages of cows and calves, and sexes of calves, were random and similar for the two groups. Calves in inbred lines were approximately 30% inbred and their dams were about 20% inbred. Outbreds were estimated to have inbreeding coefficients of 5%. Outbred matings exceeded inbred matings 30% in number of calves raised to weaning and 18% in average weaning weights. Bull calves were individually fed for 112 days. Outbred bulls exceeded inbreds 14% in yearling grade, 7% in final weight, and 3% in daily gain, but required 3% more feed per pound of gain. Differences in number raised, weaning weight, yearling grade and final weight were highly significant.

A STUDY OF THE IMMUNOGENETIC BEHAVIOR OF INBRED LINES OF HEREFORD CATTLE. C. P. Stroble, R. W. Mason and N. W. Hilston, *University of Wyoming.*

The probable blood groups of 621 Hereford cattle were formulated and studied. The cattle were from the 9 lines now living bred at the Miles City Station. IBM cards were prepared for each animal. Each IBM card contained individual identification, a code of the probable blood groups and a coded line designation. Sorts were made on a basis of blood groups in 7 of the blood group systems. From these sorts, duplicated probable blood group formulae were arranged in order of frequency of duplication within the herd. Similar sorts and arrangements were conducted on a within line basis. Differences were noted between lines both in per cent of probable blood group formulae duplicate and in the probable blood group formulae most frequently duplicated. Differences were noted between lines when studied on a within blood group system basis. The use of probable blood group formulae as a tool for selection for between line crosses is suggested.

SELECTION INDICES BASED ON THE MILK, FAT PER CENT, FAT, AND TYPE CLASSIFICATION OF DAIRY CATTLE. K. A. Tabler and R. W. Touchberry, *University of Illinois.*

The H.I.R. lactation and type records of 2,810 daughters and mates of 756 sires from 414 Jersey herds were analyzed. The lactation records were adjusted to a 305–2X–M.E. basis. The heritability of differences in single records of milk yield, fat yield, fat percent, and type were estimated by doubling the intra-sire regression of daughter on dam and were 0.25, 0.20, 0.56, and 0.25, respectively. For the daughters' records the intra-sire phenotypic correlations were —0.36 between milk yield and fat percent, 0.88 between milk yield and fat yield, 0.08 between milk yield and type, 0.15 between fat percent and fat yield, 0.05 between fat percent and type, and 0.11 between fat yield and type. The corresponding intra-sire genetic correlations were —0.50, 0.72, 0.07, 0.20, —0.01, and 0.08, respectively. Indices for estimating the genic values for milk yield, fat percent, fat yield, and type by utilizing the information on the phenotypes of these traits

were developed. The expected genetic gains of the indices indicated that the genic value for milk yield can be estimated 10% more accurately when the cow's fat yield is also considered. Fat yield alone is a good indicator of a cow's genic value for fat yield. The net gains of the indices were compared by converting the expected genetic gains in milk and fat yield to a milk equivalent basis with the result that milk yield alone was a good criterion of selection. Selection for type along with milk yield resulted in a 15% decrease in the expected genetic gain of milk and fat yield on a milk equivalent basis.

VARIATIONS IN THE BIRTH WEIGHTS OF PUREBRED AND CROSSBRED HOLSTEIN AND GUERNSEY CALVES. R. W. Touchberry and K. A. Tabler, *University of Illinois*

During the last four years the 40 purebred Holstein and Guernsey foundation cows in the crossbreeding project at the University of Illinois have given birth to 141 normal term, single birth calves as a result of reciprocal matings to each of two Holstein and two Guernsey sires. Of these calves 77 were females and 64 were males, 36 were purebred Holsteins, 35 were purebred Guernseys, 34 were crossbreds by Holstein sires and out of Guernsey dams and 36 were crossbreds by Guernsey sires and out of Holstein dams. The birth weights of these calves were fitted by least squares to a mathematical model in which it was assumed that the breed of the sire, the sires within breeds, the breed of the dam, the sex of the calf, the weight of the dam and the interactions involving these main effects were instrumental in causing variations in the birth weights. By solving the necessary equations it was found that those calves by Holstein sires were 7.3 lb. heavier than those by Guernsey sires and those out of Holstein dams were 19.3 lb. heavier than those out of Guernsey dams. The crossbreds were 2.6 lb. heavier than the purebreds as was indicated by the breed of sire by breed of dam interaction. The male calves were 5.1 lb. heavier than the females and the birth weight increased 0.5 lb. for each 100 lb. increase in the weight of the dam.

THE MINERAL CONTENT OF CATTLE HAIR. R. G. Washburn, L. O. Gilmore and N. S. Fechheimer, *Ohio Agricultural Experiment Station.*

The hair from 51 cows was analyzed for its total ash content with a range of 0.28% to 3.08%. The hair was selected to represent four colors: Black, White, Brown and Red. The average ash content of these samples was as follows: Black, 2.29%; White, 1.63%; Brown, 1.48% and Red, 1.71%. The ash from these hairs was analyzed spectrographically using Lithium Fluoride to dilute the pure ash. The proportions were 90 parts Lithium Fluoride, 10 parts ash. The ratios of the following elements were determined: Mg, P, Mn, Fe, Cn, Ag, Ca, Pb, and Zn. A flame photometer was used to determine the percent of Na and K present in the ash. The Na content averaged 1.69% and the K content averaged 2.18%. The possibility of genetic factors causing difference in the percentage composition of Ca, Pb and Zn is sugested from the available data.

HERITABILITY OF THE OPEN-FACED TRAIT IN HAMPSHIRE SHEEP. Thomas D. Watkins, Jr., *U. S. Department of Agriculture.*

A study was made of the yearling records of 35 Hampshire rams, 206 ewes, and their 446 progeny produced from 1929 through 1952 at the Agricultural Research Center, Beltsvile, Maryand. Preiminary heritability estimates for the open-faced trait as determined by the paternal half-sib correlation and the intra-sire regression methods were 0.62 ± 0.05 and 0.34 ± 0.08, respectively. These estimates were

fairly high and indicated that rapid progress could be made if selection for this were intensified in the flock at Beltsville. On the basis of a five point scoring system, significant differences were found between the mean face scores of yearling rams and ewes, and between face scores of progeny when classed by age of dam—immature, mature, and aged. Face scores in the final analysis were adjusted to the yearling ewe, mature dam basis. The above heritability estimates were corroborated by the use of components of variance and by breaking the sire group data of the many years into 2×2 and 2×3 blocks and averaging the sire component of the analysis of variance for comparison with the sire component of the analysis of variance used for determining the heritability estimates. From the average sire component of the blocks, the sire component was 0.0838 while that of the analysis of variance used for computing the heritability estimates was 0.0651. Thus it appears that the heritability estimates, although moderately high, were rather conservative.

GENETIC STUDIES OF STEER PROGENY GROUPS SLAUGHTERED FOLLOWING THREE SUCCESSIVE FEEDING TREATMENTS. Fred S. Willson, Joe Urick and Alva E. Flower, *Montana State College.*

Two hundred seventy-two steer progeny of 36 Hereford sires covering a period of eight years were group fed a limited ration their first winter, grazed on mountain meadow their second summer and full-fed a fattening ration the following winter. Daily gains for each steer were computed for these periods, designated 1, 2, and 3, respectively. Genetic correlations between the three periods were: $r_{G1G2} = 0.304$, $r_{G1G3} = 0.692$, $r_{G2G3} = 0.220$. This indicates some of the genes controlling growth each of the three periods are the same. Heritability estimates of gains made the three periods: $h_1 = 0.543$, $h_2 = 0.746$, $h_3 = 0.418$. Heritability estimates indicate selection for gains any of the periods should yield genetic progress. Genetic correlations between area of loin eye muscle and daily gain in successive periods varied from 0.79 to an estimated 1.0. Genetic correlations between fat over eye and daily gains in the suceeeding periods varied from -0.118 to an estimated -1.0. Heritability estimates of loin eye muscle and fat over loin eye were: h eye muscle $= 0.727$, h fat $= 0.583$. This indicates a negative relationship between development of red meat and fattening potential at this age. Heritabilities indicate that either can be genetically changed.

BODY MEASUREMENTS INDEXES AS A MEANS OF SELECTION IN SHORTHORN CATTLE. T. S. Yao, R. L. Hiner and W. M. Dawson, *U. S. Department of Agriculture.*

Body measurements were taken just before slaughter at 900 lb. on 287 beef and dual-purpose type Shorthorn steers at the Agricultural Research Center, Beltsville, Md. The steers were individually full fed from 500 to 900 lb. Seven body measurement indexes: (A) height at withers/width of chest; (B) height at withers/circumference at chest; (C) length of body/width of chest; (D) length of body/circumference at chest; (E) height at withers $\times$ length of body/width at chest; (F) height at withers $\times$ length of body/circumference at chest; and (G) height at withers $\times$ length of body/width at chest $\times$ depth at chest were correlated with 15 beef production characters, and with scores for slaughter grade and body compactness. Beef production characters studied were: dressing percentage; bone percentage of 9-10-11 ribs; carcass grade; average daily gain; slaughter weight; lean fat ratio 9-10-11 ribs; lean bone ratio 9-10-11 ribs; eye muscle area; and weights of head; skull; legs, carcass; 6-12 ribs; bone and tendon of 9-10-11 ribs; lean of 9-10-11 ribs. All of the measurement indexes were significantly corre-

lated with slaughter grade and body compactness. In general, they were more highly correlated with the beef production characters than the single body measurements were but not with as many as slaughter grade was. Of the measurement indexes only index B was highly heritable (45.0%). It is suggested that this index could be used advantageously as an objective measure in selection.

INHERITANCE OF BLACK FIBERS AND SKIN COLOR AND THEIR RELATION TO PRODUCTION CHARACTERS IN HAMPSHIRE SHEEP. T. S. Yao, Thomas D. Watkins, Jr., Carl G. Potts and Damon A. Spencer, *U. S. Department of Agriculture.*

The scores for black fibers in fleece and skin color of 348 Hampshire yearling sheep, and their 164 dams and 24 sires, were analyzed with five production characters—weaning weight, fleece weight, flesh condition, birth type and reproductivity of ewes. The animals having no black fibers in fleece produced 24.4% offspring having black fibers, while the animals having black fibers in fleece produced 50.0% offspring without black fibers. When one parent had black fibers, 64.5 and 67.1% of their offspring were without black fibers. Similar results were obtained for skin color. Sheep having pink skin produced 53.7% dark skinned offspring and the dark skinned animals produced 26.4% pink skinned offspring. Both characters seemed to have a number of multiple quantitative genes, which caused the appearance of the black fiber and dark skinned offspring from the animals without black fibers and of pink skin, and vice versa. The heritability estimates of black fibers were 28.5 and 25.2% from the half-sib correlation and regression methods respectively, average 26.9%. The heritability estimates for skin color were 23.0 and 37.3% from the above methods respectively, average 30.2%. There was no significant correlation between these two characters and the five production characters except betweeen the skin color and fleece production (0.123 moderately significant). The results revealed that the black fiber and dark skin characters may be improved through a special breeding project along with an intensive progeny test without seriously impairing the lamb and wool production.

Meats Section

C. H. Adams, *Chairman*

THE EFFECT OF STILBESTROL AND STILBESTROL-PROGESTERONE IMPLANTS UPON THE FEED LOT PERFORMANCE, CARCASS QUALITIES, AND REPRODUCTIVE ORGANS OF EWE AND WETHER FEEDER LAMBS. T. Donald Bell, Walter H. Smith, Andrew B. Erhart and Lewis A. Holland, *Kansas State College.*

Three lots of 48 feeder lambs each received similar rations of sorghum roughage, sorghum grain, protein supplement, limestone and salt. One-half the lambs in one lot received two pellet implants at the start of the tests. Each pellet contained 30 mg. of progesterone and 3 mg. of stilbestrol. The remaining one-half of the lambs in the lot were implanted with 4 pellets. The lambs in another lot were divided into 3 portions. One of these portions received 15 mg. stilbestrol implants; another received 6 mg. stilbestrol implants; and the third portion received 12 mg. stilbestrol implants secured from a different manufacturer. The third lot of lambs

received only the basal ration and served as controls. The hormone treated lots of lambs ate slightly more feed, and made more rapid and more efficient feed lot gains than the untreated lot. Gains were similar in all the hormone treated groups with the exception of the slightly lower gains of the lambs receiving the 6 mg. stilbestrol implants. The carcasses of the treated lambs graded one full grade lower and the yield was 4% less than the untreated lambs. Detailed carcass studies, cooking and palatability tests, and chemical analyses of tissues from representative lambs from each of the three lots revealed differences between the treated and untreated lambs. Consistent abnormal development was found in the urogenital systems of the treated lambs.

INFLUENCE OF CHANGING THE KIND OF FAT IN THE DIET AT VARIOUS WEIGHT INTERVALS ON CARCASS FAT CHARACTERISTICS OF SWINE. T. N. Blumer, E. R. Barrick, W. L. Brown, F. H. Smith and W. W. M. Smart, Jr., *North Carolina Agricultural Experiment Station.*

A basal diet containing 10% added soybean oil was fed to pigs from 10 weeks of age to weights of 125, 150 and 175 lb. The soybean oil was then replaced by an equal quantity of coconut oil and fed until the pigs weighed about 200 lb., at which time they were slaughtered. Control animals were fed each oil continuously to slaughter weight. Rate of gain and feed efficiency were very similar for all diets. Carcass firmness scores averaged firm, medium firm-firm, medium firm, medium soft and and soft for the coconut, 125, 150, 175 and soybean groups, respectively. The percent lean cuts, average back fat thickness and loin area were not appreciably different. A significant difference was found in the iodine number, content of linoleic, linolenic, saturated acids and arachidonic acid of the pork fat produced by the different fat diets. Oleic acid content was not appreciably influenced by the fat fed. Keeping quality of the fat as measured by peroxide values of the stored cured bacon samples indicated that hogs continuously fed coconut oil produced the most stable fat, followed by those fed coconut oil from 125 lb. The 150 and 175 lb. groups were superior to the soybean group in fat stability but were only slightly different from each other. The data indicate that hogs fed soybean oil may be hardened satisfactorily by substituting coconut oil in the diet at/or before a weight of 150 lb.

DEVELOPMENT OF PROTEIN AND FAT IN FATTENING STEER AND HEIFER CALVES. G. A. Branaman and Paul E. Howe, *Michigan State College and U. S. Department of Agriculture.*

The cooperative study of fattening of 36 steers and 36 heifers (1932–35) supplies data on the composition of carcasses and the quantities of protein, fat and bone developed in Hereford cattle from Miles City, Montana, Station during 300–500 days. The curve of growth is approximately straight on semi-logarithmic paper from 330 to 425 days after which it decreases in slope. Similar changes occur in the average data in record of performance studies. The slopes calculated from the values obtained by least squares and the correlation coefficient of the latter in parentheses are as follows: for 330–425 days, Weight of lean—Steers, .0030 (.81); Heifers, .0021 (.72): Weight of 9–11 rib-eye muscle—Steers, .0015 (.47); Heifers, .0013 (.33): Weight of fat—Steers, .0079 (.56); Heifers, .0037 (.50): Weight of bone—Steers, .0022 (.55); Heifers, .0014 (.67). For 426–510 days, Weight of lean—Steers, .0009 (.32); Heifers, .0008 (.04): Weight of eye—Steers, .0003 (.09); Heifers, .0002 (.05): Weight of fat—Steers, .0026 (.28); Heifers, — .0016 (— .20). Weight of bone—Steers, — .0022 (— .06); Heifers, — .0011 (— .23). These results apply to the transitional period between rapid development and fattening in ROP

studies and to the production of lean. Protein (and its associated nutrients) is the unique contribution of animals to man's diet. Environment and genetic factors have a considerable influence on development from birth to weaning with resultant difference in weight and size, which tends to persist in the test periods. Development of muscle beyond weaning is fairly uniform, the difference in rate of growth being predominantly in fat.

THE POSSIBLE FUNCTIONS OF DIETHYLSTILBESTROL AS OBSERVED IN BEEF CARCASS EVALUATION AND CERTAIN ASSOCIATED GLANDS. V. R. Cahill, E. W. Klosterman, F. E. Deatherage and L. E. Kunkle, *The Ohio State University*.

A project designed to study the effect of diethylstilbestrol implantation in bulls and steers has been carried to the completion of one cycle. Forty Hereford bull calves of similar breeding were used; twenty were castrated and divided into two lots of 10 steers each to pair with two lots of 10 bulls each. The cattle in one lot of steers and in one lot of bulls were implanted with 105 mg. of diethylstilbestrol at the beginning of the feeding trial and again 84 days later.

The data indicate that diethylstilbestrol tends to induce deposition of fat on bulls and retard it on steers and to increase the gross weight of adrenal glands in steers and the pituitary gland in both steers and bulls.

FURTHER STUDIES ON THE ANABOLIC EFFECTS OF STILBESTROL IN STEERS AS INDICATED BY CARCASS COMPOSITION. M. T. Clegg and F. D. Carroll, *University of California*.

Sixteen two-year old Hereford steers were randomly divided into two groups of eight steers each. Both groups were placed on the same fattening ration—barley, molasses dried beet pulp, cottonseed meal, alfalfa hay and grain hay. Group 1 was left untreated, and Group 2 was treated with five 12 mg. pellets (60 mg.) of stilbestrol implanted subcutaneously in the ear. These animals remained on feed for 104 days at which time they were slaughtered and a sample rib cut from each carcass analyzed for percent fat, protein, bone and moisture. In addition the percentage cut out values of the wholesale cuts were determined. After slaughter, the seminal vesicles were dissected out, weighed and fixed for histological study.

An analysis of the seven rib cut (6, 7, 8, 9, 10, 11, and 12) indicates the following: There is no difference in the percentage of moisture; a smaller percentage of fat; a greater percentage of lean; a smaller percentage of ether extract; and a greater eye muscle area in the treated animals over the controls. An evaluation of the carcass cut out percentage values substantiates the above findings. The average weight of the seminal vesicles in the treated animals was 34.0 gm. as contrasted to 8.5 gm. in the untreated group. Histological studies of the seminal vesicles showed the treated glands to contain columnar epithelial cells which give the appearance of actively secreting as contrasted to a somewhat low cuboidal type epithelial cell in the untreated group. The weight of the kidney in the stilbestrol group was significantly larger than the controls.

THE IMPORTANCE OF FAT FIRMNESS IN EVALUATION OF PORK CARCASSES. J. W. Cole, Leon Orme and Charles S. Hobbs, *University of Tennessee*.

Firty-nine carcasses were selected from a local packing plant on a basis of five degrees of firmness (hard, medium hard, medium soft, soft and oily) as measured

by a penetrometer, and further grouped according to amount of finish—underfinished, intermediate finish and overfinish. The following data were obtained: cutting test, measurements, cooking and palatability tests, freezing rate and shrink, iodine and peroxide values on rendered lard and curing losses. Degree of firmness does not influence the percentage of primal cuts or percentage of fat. The longer carcasses did yield a higher percentage of primal cuts, based on carcass weight. No differences in cooking losses could be attributed to the degree of hardness of the fat. There was no consistent preference for any roast of any one degree of finish or hardness of fat. Shearing strengths on the cooked meat fell within the range of 10.2 to 15.2 lb. Individual variations among animals seemed to have more effect than the degree of finish or firmness of the carcass fat. There is no definite trend concerning the influence of firmness or degree of finish on freezing rate and shrink. The iodine number of the various samples substantiated the penetrometer measurements. The peroxide value tends to increase with storage. Firmness seems to have some effect on this value, with the harder fat having the lowest readings. There appear to be no significant differences in curing losses between the five degrees of firmness or between the varying amounts of finish.

THE EFFECTS OF DELAYED APPLICATION OF CERTAIN DRY CURES ON THE PRESERVATION OF QUALITY OF PORK. M. C. Heck and Martin J. Burris, *University of Arkansas.*

The hams and shoulders of fifteen choice butcher hogs were randomly allotted to six treatment groups. Immediately after the carcasses were chilled and cut, the following dry cures—sugar, salt, and special (Smithfield)—were applied to cuts in Groups 1, 2, and 3 respectively, using curing practices recommended for each cure. The same cures were used for Groups 4, 5, and 6 but the application of cure was delayed for ninety-six hours after chilling. Center cut samples of hams and shoulders were cooked and a panel of judges scored the samples for aroma, flavor of fat and lean, texture, tenderness, and juiciness. Delaying application of the cure improved tenderness and texture of the sugar-cured and special-cured cuts. In addition the aroma and flavor of lean of the special-cured cuts was markedly improved by this treatment. Delaying application of the salt cure decreased the tenderness and texture scores of both cuts, decreased the juiciness of the hams and produced some improvement in aroma of hams and shoulders.

EFFECT OF PLANE OF WINTER NUTRITION ON QUALITY OF BEEF. Harold B. Hedrick, D. E. Brady and A. J. Dyer, *University of Missouri.*

Thirty-three head of yearling Hereford feeder steers were divided to give three levels of winter gain; 1.5, 1.0 and —0.4 lb. per head daily. All cattle were grazed during the summer and finished in dry lot to the Choice grade. Carcass measurements and physical, chemical and organoleptic studies of the 9–10–11th rib were made. Analysis of variance showed highly significant differences between lots in percent fat and lean in the 9–10–11th rib and percent ether extract in the rib eye. Carcasses from cattle on the low plane of winter nutrition had more separable fat, less separable lean and less fat in the rib eye than carcasses from the lots on the higher plane of winter nutrition. The palatability committee found no significant difference in tenderness between lots in cooked rib roasts and rib steaks. Shear tests corroborated the results of the panel. Analysis of variance of carcass measurements showed no significant differences between lots. Carcasses from the low plane of winter nutrition lot graded lower and showed greater grade variability than the other lots.

WASTE BEEF FAT IN STEER FATTENING RATIONS AND ITS EFFECT UPON THE CARCASS. J. F. Hentges, Jr., A. M. Pearson and Cecil A. Tucker, II, *Florida Agricultural Experiment Station.*

Two trials were conducted to determine the feasibility of using raw ground waste beef fat in steer fattening rations. In the first trial 18-month old steers of mixed breeding were fed 90 days on either a basal ration or one containing 5% added fat. The steers receiving fat gained 2.3 lb. daily compared to 1.8 lb. for the basal, and required less feed per unit of gain. However, there was little difference in carcass grade or dressing percentage. In the second trial, three lots of steer calves 8 months old were fed for 154 days on a basal ration (Lot 1), 5% fat (Lot 2), or 10% fat (Lot 3). Average daily gains were 1.8, 1.9 and 1.5 lb. for Lots 1 through 3, respectively. Lot 2 required the least feed per unit of gain and Lot 1 the most. There was no measurable difference between lots in regard to either carcass grades or dressing percentages. The percentage of separable fat in the 9–10–11 rib cut was highest for Lot 2 with the other two lots being about equal, but the difference was not statistically significant. There was little difference between lots in tenderness as measured either organoleptically or by shear strength. Results indicated waste beef fat is satisfactory in steer fattening rations up to 5% of the concentrate mixture.

A STUDY OF THE QUANTITATIVE AND QUALITATIVE CHARACTERS OF BEEF-RIB. R. L. Hiner and T. S. Yao, *U. S. Department of Agriculture.*

An analysis of certain rib-eye data from the dressed carcasses of 304 beef and dual-purpose type Shorthorn steers showed that beef-type steers had significantly higher subjective scores for: size of eye muscle, marbling of rib eye, width of eye, intensity of flavor of lean, quality and quantity of juice and organoleptic tenderness. All of the steers were slaughtered when they reached a final feed-lot weight of approximately 900 lb. The dual-purpose type carcasses had longer and heavier eye muscles in the 9–10–11 rib, and a wider ratio of length to width of eye muscle, an index of shape. Studies of four related generations indicate that in case of the dual-purpose type steers, size of eye muscle had increased, length decreased and width increased, marbling more abundant and intensity of flavor of lean more pronounced. Also length of eye muscle had decreased and intensity and desirability of lean had increased for the four generations of beef-type steers. Heritability coefficients estimated by the half-sib method, show that when the high level of heritability is set at 40%, the following factors may be effectively selected for: size of eye, length and width of eye, ether extract fat, marbling, intensity and desirability of lean, quality of juice and resistance to shearing.

CARCASS CHARACTERISTICS OF HEAVY CALVES PRODUCED BY THE KENTUCKY COW-AND-CALF PLAN. James D. Kemp, E. J. Wilford and W. P. Garrigus, *University of Kentucky.*

Carcass characteristics were determined on 72 cattle (45 steers and 27 heifers) of known history produced over a four-year period by the Kentucky Cow-and-Calf plan. They were produced from grade cows of mixed dairy and beef breeding with equal numbers being sired by purebred Angus, Hereford, and Shorthorn bulls. They grew and fattened without grain, on milk and pasture and were slaughtered within a few hours after removal from their dams. They were from 6 to 10 months of age and ranged in weight from 344 to 808 lb. and averaged 612 lb. The carcasses were graded by a USDA grader as 3 Prime, 44 Choice, 27 Good and 1 Commercial with respective dressing percentages of 58.7, 58.3, 55.9, and 54.3. The lean was

usually bright red and the fat slightly yellow. Cutting percentages showed little differences among weight, sex, grade or breed groups although calves of the higher grades showed more kidney knob and flank. Physical separation of the rib cut showed the Choice carcasses to be much fatter than the Good carcasses (34.6 to 26.8% fat) and heifer carcasses tended to be fatter than steer carcasses (33.9 to 30.2% fat). Offal weights and carcass measurements were similar among breeds except that hides from Hereford sired calves were heavier followed by Angus and Shorthorn.

A COMPARISON OF CARCASSES FROM PUREBRED BRAHMAN STEERS WITH STEERS OF VARYING PROPORTIONS OF SHORTHORN AND BRAHMAN BREEDING. A. M. Pearson, J. W. Carpenter, W. G. Kirk, Marvin Koger and D. H. Kropf, *Florida Agricultural Experiment Station.*

Thirty-one steers representing purebred Brahman, ¾ Shorthorn-¼ Brahman, ½ Shorthorn-½ Brahman, or ¼ Shorthorn-¾ Brahman were fattened on the same ration and slaughtered under identical conditions. Differences in average daily gains, dressing percentage, head weight, percentage of carcass made up by chuck and short loin were not significant. Differences in the length of carcass and circumference of round were not statistically significant, yet the length of the hind leg became progressively greater as the percentage of Brahman breding was increased. Physical separation of the 9–10–11 rib showed the cattle with ¾ Shorthorn breeding had more fat and less lean than those carrying more Brahman blood, but differences in the amount of bone were not significant. The greater amount of fat as the amount of Shorthorn breeding was increased was borne out by a corresponding increase in the percentage of kidney knob. The carcass grade became progressively lower as the percentage of Brahman breeding was increased. Tenderness ratings by an organoleptic committee showed a similar relationship to exist as the proportion of Brahman breeding became higher. However, the percentage of round was significantly higher for the purebred Brahman steers.

RELATIONSHIP BETWEEN PHYSICAL MEASUREMENTS AND THE FAT AND WATER CONTENT OF HOG CARCASSES. R. L. Skinner, J. Kastelic, E. A. Kline and P. G. Homeyer, *Iowa State College.*

The relationships between certain physical measurements and the fat and water content of carcasses and cuts from 50 hogs were investigated. Hogs weighing approximately 200 lb., selected on basis of apparent muscling and degree of finish, were divided into three groups: meat, medium, and fat. Physical measurements included live probe measurements, back fat thickness, specific gravity of the carcass and percent lean cuts. The percent fat and water in the ham, loin, picnic, boston butt, and belly from the right half and the percent fat and water in the left half of each carcass were determined. The simple correlation coefficients for all possible combinations between the fat content of the carcass, the fat content of the cuts, and the physical measurements for the combined groups of meat, medium, and fat hogs were significant. The arithmetic average of all correlations for the fat content of the half carcass and the fat content of the cuts and the physical measurements was 0.681, 0.509, 0.420 for the meat, medium, and fat groups of hogs respectively. An equation permitting substitution of the data for back fat thickness, live probe, and percent fat in the picnic ($R = 0.893$) was found to be reliable for estimating the fat content of hog carcasses. Since correlation coefficients between percent fat and percent water in the various cuts were above 0.917 and highly significant, the data for the water content can be used to determine the fat content of the cuts.

EVALUATION OF BASIC FACTORS FOR ESTABLISHING GRADES FOR SOW CARCASSES. C. L. Strong, J. C. Pierce and F. J. Beard, *U. S. Department of Agriculture.*

Measurement and cutting data were collected for 313 sow carcasses and analyzed to study relationships that could be used to characterize grades. Average back fat thickness was the measurement most closely related to yields of the four trimmed lean cuts (hams, loins, picnics, and butts), with a simple correlation of —.90. Each 0.1 inch change in average back fat thickness reflected a change of 0.77% in lean cut yield. In multiple correlation analyses, length and weight of carcass, either singly or in combination, added little to the accuracy of average back fat thickness for predicting yields of lean cuts. Analysis of data on an additional sample of 103 carcasses confirmed the relationships revealed in the original data. In order to study data representative of the usual method of handling heavy weight cuts from sows, the four lean cuts from 58 of the 103 carcasses were boned and yields of lean ham trimmings, lean picnic trimmings, boneless loin strips, and boneless butts were determined. Yields of boneless cuts from carcass weight were closely related to yields of the four lean cuts. Measurements had similar but slightly lower correlations with the boneless yields than with yields of trimmed lean cuts. The relationships determined in these data indicate that carcass measurements may be used as reliable guides to grades for sow carcasses and slaughter sows.

VARIATION IN FEED EFFICIENCY AND CARCASS CHARACTERISTICS OF INDIVIDUALLY FED SWINE. M. Vorkapich, J. A. Hoefer and L. J. Bratzler, *Michigan State College.*

Nine crossbred pigs, six of which were litter mates of a Duroc-Berkshire cross and three Yorkshire-Chester White (litter mates), five barrows and four gilts, were divided into nine lots and placed on self-feeders containing a 16% protein ration. Throughout the experiment, the pigs and the amount of feed consumed were weighed weekly. At 125 lb. the protein was reduced to 12%. The pigs were probed at three points; posterior to the shoulder blade, lumbar region, and ham, at the live weights of 100, 150 and 215 lb. which was the slaughter weight. Carcass data included primal cut weight, body and leg length, backfat and eye muscle measurements. The primal cutout ranged from 46.7 to 53.0% Average backfat measurements ranged from 1.44 to 2.11 inches. Consumption of feed per 100 lb. of gain ranged from 337 to 422 lb.

EXTENSIBILITY OF SINGLE BEEF MUSCLE FIBERS. Hsi Wang, D. M. Doty, F. J. Beard, J. C. Pierce and O. G. Hankins, *American Meat Institute Foundation, The University of Chicago, and U. S. Department of Agriculture.*

For this work, 43 *Longissimus dorsi* and 13 *Semitendinosus* samples, some aged at 35° F. for 2 and 4 weeks, from 15 carcasses (3 each of lightweight prime, heavy prime, lightweight good, heavy good and commercial cow) were used. Muscle fibers were isolated by cutting $2 \times 3 \times 0.5$ cm. pieces of meat in a Waring Blendor. Single muscle fibers were placed in water in a Petri dish with a ruler beneath it. Using No. 1 objective and $5\times$ oculars on a binocular microscope, a single muscle fiber was gripped tightly with two steel forceps leaving a distance of 5 mm. between them. One of the two forceps was pulled until the fiber broke. The length of the stretched fiber at the breaking point was noted and the increase in length recorded as "extensibility." Twenty such readings were made for each sample and their arithmetic mean calculated. The influence of aging on the extensibility of raw and cooked muscle fibers was not consistent for *Longissimus dorsi*

samples, but a substantial and consistent decrease in extensibility was noted in the *Semitendinosus* samples, especially the cooked. On the other hand, cooking always increased the extensibility of muscle fibers: to a greater extent in good grade and commercial cow than in prime grade samples, and in samples from heavy than from lightweight carcasses of corresponding grade. The extensibility of raw muscle fibers did not appear closely correlated with shear strength of either the raw or cooked tissue, or tenderness score. Extensibility of cooked muscle fibers was, however, significantly related to shear strength of the cooked meat and to tenderness score in 3 out of 5 carcass classes (32 *Longissimus dorsi* samples), but not in the remaining 2 carcass classes (11 *Longissimus dorsi* samples).

BODY COMPOSITION AND CARCASS CHANGES OF YOUNG CATTLE. G. H. Wellington, J. T. Reid, L. J. Bratzler and J. I. Miller, *Cornell University*.

The effect of age increases and of three levels of nutrient intake on the amount of edible meat, carcass length and thickness, weights of individual muscles, and tenderness were analyzed. The accuracy of body water composition determined *in vivo* by intravenous injection of a known amount of antipyrine and measuring the degree of dilution of the drug in all body water was evaluated. The percentage of body fat was calculated from the amount of body water. Cattle with greater T.D.N. intake had a higher dressing percentage, increased length and thickness of carcass and a larger ratio of edible meat to bone. Higher nutritional levels were associated with a smaller percentage weight of muscles in the carcass. The level of feeding tended to show no significant influence on tenderness. As the cattle increased in age there were significant increases in length and thickness of the carcass and a larger ratio of edible meat to bone. Age showed no influence on dressing percentage and in general no consistent influence on percentage weight of muscles in the carcass. The total body water calculated for twenty cattle by the antipyrine technique was highly correlated, .939, with the total body water determined by analyzing all body tissues for moisture.

POST-MORTEM CHANGES IN MUSCLE PLASMA PROTEINS. Eugen Wierbicki and F. E. Deatherage, *Ohio Agricultural Experiment Station*.

Recent investigations have indicated that muscle plasma proteins are closely related to tenderness and shrink in meat. Post-mortem formation of actomyosin seems to be related to the initial toughening of meat. There are indications that actomyosin may dissociate to actin and myosin to a very limited extent but evidence points to the possibility of the ions on the actomyosin-molecule may change during aging in such a way as to cause changes in the degree of hydration. Such post-mortem changes in the ion protein interrelations may account for tenderization on aging. Shrinkage of meat on cooking and drip on freezing may also be related to these ion protein interactions.

THE EFFECT OF ASCORBIC ACID AND PACKAGE DESIGN ON THE RETENTION OF COLOR IN BOLOGNA. G. D. Wilson, *American Meat Institute Foundation*.

In three replicate experiments bologna was prepared containing ascorbic acid and sodium ascorbate and the initial color and rate of fading of these products were compared to a control containing no additives. Bologna from each lot was packaged in cellophane and vacuum packages and stored at 34° and 40° F. to determine the effect of these variables on color retention. Color was measured visually and spectrophotometrically. Considerable variation in the initial color of

the treated lots was observed but generally the addition of ascorbic acid or its salt resulted in a slightly improved initial color and some prolonging of the fading process. Vacuum packaging increased the shelf life of the product in all experiments but the results indicate that some processing changes are required to obtain maximum protection in flexible vacuum packages. Packaged bologna stored at 34° F. retained its natural color longer than a similar product stored at 40° F. The lower storage temperature was also effective in retarding bacterial spoilage.

Nutrition Section

W. E. Dinusson, *Chairman*

UTILIZATION OF PHOSPHORUS FROM VARIOUS INORGANIC SOURCES BY STEERS. C. B. Ammerman, A. L. Neumann, R. M. Forbes and H. W. Norton, *University of Illinois.*

By using the balance technique, yearling steers averaging approximately 550 lb. at the beginning of the experiment were used for evaluating various inorganic phosphates as a source of phosphorus. Supplements included in the test were two dicalcium phosphates, bone-meal, defluorinated rock phosphate, imported rock phosphate and colloidal phosphate. Rations consisted of prairie hay, cerelose, corn gluten meal, corn oil, urea (which provided 55% of the total nitrogen), trace minerals and the various supplements studied. The total phosphorus intake was maintained at a level considered to be just suboptimal for good gains. Approximately one half of the total phosphorus was supplied by the supplement in question and the other half by the basal ration. By the addition of calcium carbonate, the Ca:P ratio of all rations was kept at 2.5:1. The design of the experiment was a 6×6 Latin Square with each period consisting of a twenty-one day preliminary feeding period followed by a seven day collection period. Only the feces were collected and analyzed for phosphorus since it was found that the urine did not contain appreciable amounts. Blood plasma phosphorus levels were determined following each collection and remained within the normal range for all steers throughout the entire experiment. Differences in percent of phosphorus retained, between phosphorus supplements tested, between steers and between periods, were not significant.

PANTOTHENIC ACID REQUIREMENT OF WEANLING PIGS FED A PURIFIED RATION. C. E. Barnhart, D. V. Catron, L. Y. Quinn and G. C. Ashton, *Iowa State College.*

Forty-eight cross-bred pigs were allotted to 12 different ration treatments to determine the pantothenic acid requirement of weanling pigs. The pigs averaged 25.4 lb. at the start of the experiment and were fed to a final weight of approximately 100 lb. They were confined to wire floored individual crates for the entire feeding period. A purified ration with levels of 2, 3, 4, 5, 6, and 7 mg. of pantothenic acid per lb. of ration with and without chlortetracycline was fed. The average daily gain and feed required for 100 lb. of gain for the pigs receiving the above levels of pantothenic acid without chlortetracycline were 1.18, 1.31, 1.18, 1.22, 1.34 and 1.28; 244, 254, 254, 248, 241 and 238 lb. respectively and with chlortetracycline 1.32, 1.30, 1.30, 1.37, 1.31 and 1.29; 221, 237, 238, 235, 230 and 229 lb.

respectively. There were no significant differences in the rate of gain, daily feed consumed, or feed required for a pound of gain between the pigs being fed the different ration treatments. Neither were there any significant differences in hemoglobin, red blood cell count, white blood cell count, differential white blood cell count, hematocrit, or clotting time between the pigs on the 12 ration treatments. The amount of calcium pantothenate excreted when the pigs weighed 75 lb. was found to be closely related to the level of pantothenic acid the pigs were receiving in their ration. No pantothenic acid deficiency symptoms were observed in any of the pigs.

EFFECTS OF AUREOMYCIN WHEN FED IN A CALF STARTER, IN MILK, OR BY CAPSULE ON THE GROWTH OF DAIRY CALVES. E. E. Bartley, F. C. Fountaine, J. J. Radisson and F. W. Atkeson, *Kansas State College.*

Five groups of 8 calves each were fed 350 lb. whole milk, alfalfa hay and a calf starter during the first 8 weeks of life. The hay and starter were fed *ad libitum* beginning at 1 week of age. Calves of the various groups were furnished Aureomycin in the following ways: none, 1% Aurofac 2A in calf starter, 45 mg. Aureomycin (from Aurofac 2A) per calf once daily by capsule commencing at birth, 45 mg. Aureomycin (from Aurofac 2A) per calf once daily in the milk commencing at birth, 125 mg. Aureomycin (from Aurofac 10) by capsule at birth and once weekly thereafter. Calves were weighed at weekly intervals. The 8th week weights in percent of birth weights were as follows: control, 176; Aureomycin in starter, 185; 45 mg. Aureomycin daily by capsule, 191; 45 mg. Aureomycin daily in milk, 207; 125 mg. Aureomycin once weekly by capsule, 183. These results indicate that calf growth may be improved when a calf starter supplies the only source of Aureomycin to young calves, but greater improvement may be obtained if the antibiotic is provided by daily additions to the milk from birth. Also it appears that when Aureomycin is fed in milk where it presumably goes directly to the abomasum the growth response is greater than when the antibiotic is fed by capsule and presumably reaches the rumen first.

FACTORS IN THE FORMULATION OF A SEMI-SYNTHETIC DIET FOR AMINO ACID STUDIES WITH THE PIG. D. E. Becker, M. C. Nesheim, S. W. Terrill and A. H. Jensen, *University of Illinois.*

A diet formulated with dextrose (25%), dried skim milk, zein, corn starch, woodflock, corn oil, methionine, histidine, isoleucine, ammonium citrate, minerals, vitamins and an antibiotic was designed to be low in lysine and tryptophan. Weanling pigs weighing about 20 lb. were individually fed. With 18% dried skim milk and 5.0% zein (1.99% nitrogen) in the diet pigs gained at a satisfactory rate and efficiency and did not differ in growth response to l-lysine levels as low as 0.54% of the diet. With 12.0% dried skim milk and 7.5% zein (2.17% nitrogen) in the diet the pigs gained at a suboptimum rate and efficiency and failed to exhibit differences in growth rate to levels of l-lysine as low as 0.37% of the diet. However, pigs fed 0.10% or more l-tryptophan exhibited faster gains and a significantly greater feed intake than pigs fed 0.06% l-tryptophan. A digestion trial revealed that the apparent digestion coefficient of the zein was 44.8% so that the latter diet, particularly, was low in digestible protein. Also pigs were noted to occasionally exhibit a sudden lack of locomotor coordination, although they were otherwise normal. Usually the pigs showed a quick recovery, but several pigs died suddenly while on test. Autopsy immediately after death revealed a severe distention of the stomach and intestines with gas. Contents of the stomach and intestines were frothy and possessed an odor of alcoholic fermentation. No pathogenic

organisms were isolated, however, yeast organisms were present and active in the stomach and cecum. Chronic or acute symptoms were noted consistently in pigs fed the diet modified to contain 50% dextrose. These observations may indicate the desirability of a lower amount of available carbohydrate in such a diet.

SOME ASPECTS OF BULK IN NON-RUMINANT NUTRITION. J. M. Bell, *University of Saskatchewan.*

Various levels of bran, alfalfa, wheat straw, beet pulp, oat hulls, corn cobs and cellulose were added to low-fibre basal rations and fed to mice in growth and digestion trials. Chemical analysis, energy determinations and bulk measurements at different stages of digestion were used to evaluate the effects of bulk and/or "fibre" in the diet on animal performance. Marked differences were noted between "bulk sources" with bran, alfalfa and beet pulp being tolerated at high levels without adverse effects on growth. Increasing levels of fibrous ingredients did not markedly affect gains or weight of feed consumed until digestible calories/gm. of dry matter became lower than 3.2. At lower digestible energy levels the nutritive value of a non-ruminant ration may be difficult to describe adequately by usual chemical methods.

THE USE OF UREA AND STILBESTROL IN RATIONS FOR FATTENING YEARLING STEERS. M. C. Bell, R. L. Murphree and C. S. Hobbs, *University of Tennessee.*

Thirty-two yearling steers averaging 696 lb. were divided into four lots of eight steers each on the basis of weight and grade. These steers were used in a feeding study for 84 days. The basal ration for Lot I consisted of grass hay, ground corn, cane molasses, cottonseed meal and minerals. Lot II was fed 50% as much cottonseed meal with sufficient urea and corn fed to give equivalent TDN and crude protein. Lot III was fed urea and corn to replace all of the cottonseed meal in the basal ration with approximately the same amount of TDN and crude protein as the first two lots. Lot IV was started on the same ration as Lot III except that each steer was fed 10 mg. of stilbestrol per day. Due to increased appetite, the steers in Lot IV were fed more than the steers in the other lots. Urea feeding to Lots II, III, and IV amounted to 17%, 34%, and 32% of the nitrogen fed to each of the respective lots. The average daily gain for the steers in Lots I, II, III, and IV was 2.05 lb., 2.12 lb., 1.76 lb., and 2.11 lb., respectively. The steers in Lot III gained less than those in the other lots; however, these differences were not statistically significant. No undesirable side effects were noted from feeding stilbestrol; however, the length of teats on the stilbestrol-fed steers was 2 to 4 times the teat length of the steers in the other lots. Only slight differences were noted in average carcass grades and dressing percentages between the lots. A mixture of urea and corn was effective in replacing one-half of the cottonseed meal in this study and in the presence of stilbestrol it was effective in replacing all of the cottonseed meal.

THE NUTRITIVE VALUE OF ACETATE, PROPIONATE, AND LACTATE FOR LAMBS. Orville G. Bentley, T. V. Hershberger, L. E. Kunkle and D. S. Bell, *Ohio Agricultural Experiment Station.*

Lambs were fed sodium acetate or propionate (45 gm. per day) as an energy supplement to a ration composed of 0.05 lb. ground corn and 0.25 lb. of a cerelose-urea mixture and a full feed of mixed clover hay. The lambs fed the acids gained slightly faster (0.02 lb. per day) than the control lot and for each pound of acid

consumed per 100 lb. of gain the acids appeared to replace 1.03 lb. of corn, 0.62 lb. of urea supplement, and 5 lb. of the hay. In a second experiment an attempt was made to compare acetate and propionic acids, lactic acid, and corn in rations which were fed as nearly isocalorically as possible. All lambs received 0.75 lb. of ground corn daily and a full feed of good quality alfalfa hay. The control lot on this ration gained 0.30 lb. per day (84 days). Other lots of lambs received an average of 40 gm. daily of either a mixture of acetate and propionate or lactate or enough ground corn to supply the same caloric intake (allowing a 50% loss during digestion for the corn) gained 0.32, 0.36 and 0.34 lb. per day. In terms of feeding value per 100 lb. of gain, one pound of lactic acid replaced 2.75 lb. of corn, while the acetic-propionic acid mixture replaced 1.36 lb. Lambs fed the acids ate as much hay as the control lambs. Volatile fatty acid analyses made on samples of rumen contents and urine gave essentially normal values. Carcass data indicated that the lambs fed the acids had slightly higher finish.

THE EFFECT OF AUREOMYCIN UPON UTILIZATION OF GRADED LEVELS OF ALFALFA BY SWINE. Verle R. Bohman, James E. Hunter and John McCormick, *University of Nevada.*

Seventy-two Minnesota #1 X Duroc weanling pigs were allotted to 8 groups according to weight, breeding and sex. These animals were fed four levels of alfalfa meal (0, 10, 30, and 50% of the ration) with and without aureomycin (10 mg./lb. of feed) in a balanced, pelleted ration with the protein equalized. All pigs were slaughtered as soon as possible after they reached 200 lb. body weight and carcass data and organ weights were obtained. As the level of alfalfa increased in the ration, the rate of gain, dressing percentage, depth of back fat, and the percent of bacon belly and fat back in the carcass significantly decreased while the percent of shoulder, ham and loin in the carcass and the weight of the stomach and large intestine significantly increased. The pigs fed aureomycin gained significantly faster with less feed per pound of gain than the controls. None of the treatments had any effect upon the carcass length, number of ascarids, or weight of the spleen, liver, small intestine or heart. These data indicate that hogs may use up to 50% alfalfa in their rations and produce a leaner carcass than control pigs fed a conventional ration. Swine apparently adjust to a high level of alfalfa intake by enlarging the size of their digestive tract where limited transitory feed storage takes place which is indicated by the increased weights of the stomach and large intestine.

HIGH-PROTEIN CORN FOR FATTENING AND WINTERING BEEF CATTLE. James Bond, A. L. Neumann, R. M. Forbes and H. W. Norton, *University of Illinois.*

One wintering and three fattening trials were conducted to study the value of a special strain of corn selected for high-protein content. In Trial I (1951–52), three lots of yearling steers were fed as follows: Lot 7, shelled corn, corn silage; Lot 8, shelled corn, corn silage, soybean oil meal; Lot 9, high-protein corn, corn silage. The rations of Lots 8 and 9 and all subsequent trials were equalized with respect to protein content. The average daily gains for 175 days were 1.81, 2.20, and 2.24 lb., respectively. Trial II (1952–53), two lots of yearling steers were fed 150 days as follows: Lot 4, shelled corn, soybean oil meal, corn silage; Lot 5, high-protein shelled corn, high-protein corn silage. The average daily gains were 2.52 and 2.40 lb., respectively. Trial III (1953–54), two lots of yearling steers were fed 200 days as follows: Lot 9, shelled corn, soybean oil meal, legume hay; Lot 10, high-protein shelled corn, legume hay. The final 26 days all steers received

an extra pound of soybean oil meal daily. Lots 9 and 10 made gains of 2.45 and 2.29 lb., respectively. In 1952, two lots of calves were wintered 160 days. For 56 days, Lot 6 received corn silage plus 1 lb. soybean oil meal daily, and Lot 7 received high-protein corn silage. The resulting average daily gains were 1.79 and 0.86 lb., respectively. For 104 days 1 lb. ground shelled corn was added to the high-protein ration to equalize the energy content. The gains were 1.66 and 1.46, for Lots 6 and 7.

THE TOXICITY OF SOME ARSENICALS FED TO GROWING-FATTENING LAMBS. L. L. Bucy, U. S. Garrigus, R. M. Forbes and W. W. Moore, *University of Illinois.*

Previous work at Illinois showed that 3-nitro-4-hydroxyphenylarsonic acid, arsanilic acid, and potassium arsenite fed, in a practical ration, to growing-fattening lambs up to 0.024% of the total ration did not affect growth or cause pathological damage. Two experiments were conducted to study the toxicity of these arsenicals. Arsonic acid was fed at 0.05, 0.1, and 0.2, and 0.4% of the total ration and the other arsenicals at levels of equal arsenic content. Weight gains were closely related to feed consumed rather than arsenic consumption. The arsonic acid rations were the most palatable while the potassium arsenite rations were least palatable. Some of the lambs on the higher levels of arsanilic acid became convulsive. Chemical analysis of the livers, kidneys, and muscles showed that all levels of arsenic supplementation caused a highly significant increase in arsenic content of all the tissues. (As p.p.m. fresh tissue, liver 5–13, kidney 2–12, muscle .40–3.50). Blood studies showed the hemoglobin, red cell, and white cell counts to be below normal and the bilirubin to be higher than normal, especially in the lambs receiving potassium arsenite and the 0.4% arsanilic acid. Histological studies showed severe liver and kidney damage. Liver abnormalities noted were: cloudy swelling, positive ionic iron reaction, phagocytes, and cirrhosis. The kidneys contained albumin casts in the collecting ducts, intracellular ionic iron in the tubule cells, and tannish intravascular pigment in the medulla. Arsonic acid appeared to be less toxic than arsanilic acid or potassium arsenite.

ORAL ADMINISTRATION OF DIETHYLSTILBESTROL FOR GROWTH AND FATTENING IN BEEF CATTLE. Wise Burroughs, C. C. Culbertson, Joseph Kastelic, Edmund W. Cheng and William H. Hale, *Iowa State College.*

Following favorable initial cattle feeding trials in which diethylstilbestrol was administered orally, it appeared desirable to test the value of this hormone material given orally in several different types of cattle rations and with a variety of different aged animals of varying feeding histories as well as with animals of different sex. Graded levels of diethylstilbestrol were fed in 5 of 6 feeding trials conducted. The rations fed in the different experiments ranged from high-corn fattening rations to high-roughage wintering rations and in one experiment the ration was approximately half grain and half roughage fed over an 8 month period. Liveweight gains were stimulated in every feeding trial where stilbestrol was placed in the ration. This stimulation in gain was greatest in the high-grain rations and lowest in the high-roughage rations, the average stimulation in liveweight gains for all rations being 20%. Feed requirements per unit of gain made by cattle was likewise reduced, averaging 11% for the 6 trials. Heifers responded approximately similar to steers and 600 lb. cattle responded as well as animals double this weight. Upon slaughtering cattle fed stilbestrol, carcass yield was unaltered from that observed with cattle receiving no stilbestrol. Carcass grade in 3 experiments was unaltered and in 2 experiments carcass grade was

reduced slightly or an amount equal to one third of one grade. Edible and inedible cattle tissues collected at the time of slaughter revealed no detectable stilbestrol residues present in tissues provided the hormone-material was removed from the ration a short time prior to slaughter.

THE EFFECT OF PELLETING RATIONS OF VARIED QUALITY ON FEED UTILIZATION BY LAMBS. H. A. Cate, J. M. Lewis, R. J. Webb, M. E. Mansfield and U. S. Garrigus, *University of Illinois.*

This study compares three rations of varied quality when self-fed to lambs. Six lots of 50 lambs each were used, one lot getting the ration as meal and the companion lot getting the same ration as pellets. The rations used were (a) alfalfa meal and ground yellow shelled corn; (b) timothy meal, ground yellow shelled corn, soybean oil meal, and molasses formulated to approximately equal ration (a) in TDN and C.P.; and (c) timothy meal and ground yellow shelled corn. Pelleting significantly increased average daily gain and feed consumption on the two rations using timothy meal as the roughage. No significant increase was shown in daily gain and feed consumption for pelleting the ration in which alfalfa meal was used as the roughage. Economy of feed utilization was better for all pelleted lots with the greatest advantage occurring as the quality of the ration decreased. Rapidity of finish was greatest on pelleted rations. Higher carcass grades were obtained on the pelleted timothy rations when compared to timothy meal, but little difference was shown in the alfalfa ration. The two lots getting the pelleted timothy rations outgained the lot getting the pelleted alfalfa ration. This would indicate satisfactory gains can be obtained on low quality roughages in self-fed lamb rations if the daily feed consumption is high. This trial indicated that pelleting alfalfa meal and corn was of slight value. The pelleting of rations containing timothy meal significantly increased consumption, economy of gain, and rate of gain.

A COMPARISON OF PHOSPHORUS FROM DIFFERENT SOURCES FOR GROWING-FINISHING SWINE. H. L. Chapman, Jr., J. Kastelic, G. C. Ashton and D. V. Catron, *Iowa State College.*

Two replicated lots of four pigs each were fed 18 ration treatments in concrete drylot to determine: (1) the relative utilization of colloidal clay with soft phosphate, dicalcium phosphate and steamed bone meal as phosphorus supplements for growing-finishing swine; (2) the effect of including these supplements at two different levels in the ration; (3) the ability of the growing pig to utilize phosphorus from plant sources and (4) the effect of chlortetracycline (aureomycin HCl) upon the utilization of phosphorus by the pig. Evaluating criteria were rate of gain, feed efficiency and blood serum phosphorus at 100 lb. of body weight. These criteria, plus breaking strength, ash and phosphorus content of femurs were used when the pigs reached 200 lb. When compared with steamed bone meal and dicalcium phosphate, the colloidal clay with soft phosphate produced significantly decreased rates of gain, feed efficiency and breaking strength of femurs. There were no significant differences in the phosphorus content of bone ash or blood serum. Growing-finishing swine did not utilize plant phosphorus as efficiently as inorganic phosphorus, as evidenced by a significantly increased feed efficiency when the phosphorus from the three supplements increased from 0.18 to 0.30% of the ration (0.50% phosphorus total). There was also a significant linear effect on gains and feed efficiency when phosphorus from plant sources decreased from 0.50 to 0.35 to 0.20% of the total ration while inorganic phosphorus was increased to maintain 0.5% phosphorus in the total ration.

IRON AND COPPER STUDIES WITH DAIRY CALVES. Cecil Conley, Gennard Matrone, G. H. Wise and R. K. Waugh, *North Carolina Agricultural Experiment Station.*

Twenty-seven male Holstein calves were fed throughout the experiment a basal diet consisting of whole milk supplemented with cobalt, magnesium and vitamins A and D. Three replications of a 3×3 factorial design, in which 0, 30 and 60 mg. of iron and 0, 3 and 6 mg. of copper were administered daily, were employed. In the first replication and supplemental regime was initiated within several days after birth, whereas in the second and third replications the calves were rendered anemic, 5–6 gm./100 ml. blood, before adding either iron or copper to the diets. Apparently a daily intake of 60 mg. of iron, starting the second day from birth, is sufficient to maintain normal concentrations of hemoglobin and of serum iron in growing calves up to 600 to 800 lb. in weight, but is not adequate to increase hemoglobin and serum iron concentrations of anemic calves to normal levels. Six mg. of copper fed daily to calves were sufficient either to maintain a normal level or to increase the serum copper concentration from a low level to normal. Under the conditions of this experiment, the levels of copper intake investigated affected neither hemoglobin nor serum iron concentrations, and the level of iron intake did not influence serum-copper concentrations.

THE EFFECT OF MOLYBDENUM UPON THE METABOLISM OF BONE. G. K. Davis and R. W. Lindenstruth, *Florida Agricultural Experiment Station.*

The toxic syndrome caused in animals by excess molybdenum includes symptoms of abnormal bone formation or development. Conditions observed have been beaded ribs in calves, stiffness, eroded and enlarged joints in cattle and crooked front legs in rabbits. Histological sections were prepared of bones from rabbits which had received toxic levels of molybdenum. Molybdenum was fed as $NaMoO_4 \cdot 2H_2O$ in a natural ration to weanling rabbits or a solution of the molybdate was administered by intraperitoneal injection. Both methods of administration produced a typical syndrome of molybdenum toxicity. Sections of the rib and radius of affected rabbits exhibited definite dystrophic changes. The normal bone architecture was destroyed and irregular patterns of bone structure were apparent. The abnormality was present in the bones from rabbits which had received molybdenum by either method. The mechanism by which molybdenum interferes with bone metabolism is unknown, although evidences of a changed phosphorus metabolism have been observed.

THE UTILIZATION OF MOLASSES BY GROWING DAIRY HEIFERS. R. F. Davis, G. W. Trimberger, K. L. Turk and J. K. Loosli, *Cornell University.*

Growth studies involving 104 dairy heifers averaging approximately 730 lb. in weight at the beginning of the experiments have been conducted over a 2 year period (1952–1954). The control ration was 20 lb. per day corn silage, approximately 10 lb. early cut grass hay, 4 lb. corn meal, and soybean oil meal to provide sufficient protein. In the experimental rations the TDN from 4 lb., 2 lb., and 1 lb. of ground corn was replaced by TDN from molasses fed at the rates of 6 lb., 3 lb., and 1.5 lb. per head per day. Digestible protein was maintained at the level of the control ration by varying the proportion of soybean oil meal and corn in the ration except during the second year one lot was fed urea and one lot received no supplemental protein. Balance studies involving 12 dairy steers averaging about 450 lb. in weight were conducted on all rations. Rations containing high levels of molasses consistently showed decreased utilization of crude protein both from natural sources and from urea. Average daily gains on the rations

fed for both years were: control ration 1.4 lb., ¼ molasses, ¾ corn, 1.47 lb., ½ molasses ½ corn 1.43 lb., and for 6 lb. molasses 1.50 lb. The rations including no protein supplement and urea, used only the second year, provided daily gains of 1.33 lb. and 1.49 lb. respectively. It was concluded that molasses is an excellent feed for growing dairy cattle when included in a ration containing good quality roughage and ample protein. This is in contrast to the results of previous work reported from this station in which the rations were based on late cut hay.

UNIDENTIFIED GROWTH FACTORS IN SWINE NUTRITION. J. R. De Zeeuw, W. M. Reynolds, G. P. Pendergrass and H. G. Luther, *Chas. Pfizer & Co. Inc.*

Several experiments with baby pigs and growing and fattening swine were conducted to study the growth promoting value of the unknown factor(s) present in one particular fermentation source (P.F.P.). Baby pigs, removed from the sow at one week of age, were reared on a practical dry feeding regime suitably supplemented with Terramycin. The milk replacer fed to four weeks of age was constructed essentially around milk and soy products. From four weeks to eight weeks of age the baby pigs were fed a starter composed principally of corn oats, and soybean oil meal. In one trial a 12% improvement in the average daily gain was elaborated by P.F.P. (0.536 lb./day vs. 0.478 lb./day). When P.F.P. was added to the prestarter and starter modified to contain 5% fish meal, a 10% response was produced (0.615 lb/day vs. 0.537 lb./day). Increases in growth response were obtained in baby pigs receiving P.F.P. in creep feed while suckling on the sow. In the growing and fattening studies, self-fed swine raised in dry lot received a complete mixed feed based on corn and soybean oil meal. A 15–20% improvement in the average daily gain to market weight was experienced with P.F.P. supplemented the all-vegetable ration (for example, 1.75 lb./day vs. 1.43 lb./day). An approximate 10% improvement in the average daily gain was observed when P.F.P. was added to a corn-soy-fish meal basal. (1.70 lb./day vs. 1.51 lb./day).

RUMEN INOCULATION STUDIES WITH CATTLE AND SHEEP. A. J. Dyer, M. E. Muhrer and W. H. Pfander, *University of Missouri.*

The effect of inoculating ruminants with rumen microorganisms at the time of ration changes and following antibiotic administration was studied. Nine yearling steers given two 100 ml. inoculums (obtained from a steer grazing fescue) before they were taken to pasture gained at the same rate as the controls. Steers that received 500 ml. inoculums every other day for the first fourteen days on wheat pasture gained at a slightly faster rate than the control steers but did not maintain their advantage. One lamb from each of 23 sets of twins received a 10 ml. inoculum weekly for 7 weeks. The inoculated lambs gained an average of 21 lb. and the control lambs 19.6 lb. Trios of fistulated wethers were given 1 million units of penicillin. One wether in each trio served as a control, one was given lypholized rumen contents, and one received fresh rumen inoculum. The inoculated sheep regained normal rumen activity a little faster than the controls. Yearling wethers in thin condition on wheat pasture received daily inoculums of 50 ml. fresh rumen material. During the first two weeks on pasture the average gains were nine and six lb. for the inoculated and control groups respectively. The weight advantage was maintained for two months. However, in other trials little benefit has been obtained from the use of dried rumen organisms with various rations.

PROTEIN LEVELS FOR PIGS IN THE SUB-TROPICS. Irwin A. Dyer and Antonio Velasquez, *Centro Nacional de Agronomia, El Salvador.*

Five groups of 9 Hampshire weanling pigs each were self-fed in drylot to approximately 80 lb. liveweight. Groups 1–5 were fed rations composed of 20, 18.5, 17, 15.5 and 14% crude protein, respectively. Each ration was composed of ground white corn, an equal mixture of shark meal and cottonseed meal as the protein supplement, rice polish, dried skim milk, bonemeal, salt, and fortified with recommended levels of vitamins and penicillin. The average daily gains and feed per pound gain for groups 1–5 were 1.02, 3.05; 1.21, 2.86; 1.29, 2.65; 1.48, 2.41; and 1.13, 3.02, respectively. The rate of gain of pigs consuming the 15.5% protein ration was significantly faster than that made by pigs fed the 20% ration. The efficiency of feed utilization was also highest with the 15.5% protein level and lowest when the 20% level was fed.

THE PROTECTIVE EFFECT OF MINERALS ON STOMACH WORM INFECTION FOR LAMBS ON PASTURE. R. J. Emerick, W. Bemrick, R. Shumard, A. L. Pope, C. A. Herrick and P. H. Phillips, *University of Wisconsin.*

Six lots of lambs initially infected with 20,000 *Haemonchus contortus* larvae were supplemented with various salt and mineral mixtures to determine the protective effect afforded sheep under pasture conditions favorable to reinfection. Lots 1, 2, and 3 received supplements of iodized salt, trace mineralized salt, and trace mineralized salt plus dicalcium phosphate respectively. These supplements with the addition of 10% phenothiazine were accessible to lots 4, 5, and 6 respectively. Weight gains, hemoglobin values, and fecal egg counts were used as criteria for measuring the response of the lambs to the infection. The animals which received iodized salt developed a severe anemia and 20% of the lambs succumbed to the infection. The fecal egg counts reached an average peak of 10.9 million eggs per lamb per day. When trace mineralized salt was fed, the fecal egg counts reached a peak of 26.9 million eggs per lamb per day thus making possible a more rapid reinfection. Under these conditions a very severe anemia was observed, and 80% of the lambs succumbed to the infection. When dicalcium phosphate was added to the trace mineralized salt, the fecal egg count reached a peak of only 6.4 million eggs per lamb per day, the severity of the anemia was reduced, and no deaths occurred. The addition of phenothiazine to the salt mixtures in each instance reduced the fecal egg counts, increased the hemoglobin values, and greatly increased the survival rate of the lambs.

FURTHER STUDIES OF THE INFLUENCE OF ADDED CARBOHYDRATE (CERELOSE) ON THE NITROGEN METABOLISM OF STEERS. J. P. Fontenot, W. D. Gallup and A. B. Nelson, *Oklahoma Agricultural Experiment Station.*

The effect of additional carbohydrate on digestion and nitrogen utilization in beef steers fed wintering-type rations was studied in two experiments. In both experiments the basal rations were composed of prairie hay, cottonseed meal, and minerals, and contained approximately 10% protein in Experiment 1, and approximately 12% in Experiment 2. Each basal ration was fed alone and supplemented with 700 and 1050 gm. cerelose. Nitrogen retention was significantly increased by cerelose additions to the 10% ration; it was slightly but not significantly increased by cerelose additions to the 12% ration. Increasing the amount of cerelose from 700 to 1050 gm. per day produced no significant effect on nitrogen retention, regardless of the protein content of the basal ration. Cerelose added to either of the basal rations resulted in a significant increase in estimated biological value of the protein. Increasing the cerelose from 700 to 1050 gm. resulted in a small but

significant increase in biological value of protein in the 12% ration, but not in the 10% ration. Cerelose additions generally resulted in significant decreases in the apparent digestibility of crude protein and crude fiber and in a significant increase in the apparent digestibility of nitrogen-free extract.

SOURCES OF UNIDENTIFIED FACTORS FOR THE PIG. D. I. Gard, S. W. Terrill, D. E. Becker, H. W. Norton and A. V. Nalbandov, *University of Illinois.*

Five possible sources of unidentified growth factors were investigated. In Tests 1 and 2 a 3% grass juice concentrate, 3% condensed menhaden fish solubles and 5% dried whey-product were studied singly and in all possible combinations with a semi-purified and with a practical diet. In Test 3 a single level of supplemental crude fiber from either dehydrated alfalfa meal or woodflock was studied in the presence or absence of a streptomycin residue with a practical diet. No antibiotic was included. In Test 1 the pigs used were raised by sows maintained on semi-purified diets; in the other tests the pigs were raised by sows receiving complete rations on legume pasture. Pigs were randomized to treatments from litter outcome groups. Pigs in Test 1 were individually-fed *ad libitum* on screens; in Tests 2 and 3 they were group-fed *ad libitum* on concrete. Grass juice concentrate significantly ($P < 0.01$) improved growth and increased feed consumption. Growth and feed intake were significantly less ($P < 0.05$) for pigs fed either fish solubles or dried whey-product than for pigs fed neither or both. Ten percent dehydrated alfalfa meal depressed growth but equivalent woodflock crude fiber failed to influence gains. Streptomycin residue at a 1.8% level promoted growth. The grass juice concentrate was tested for estrogenic activity by the uterine weight method using one-month old castrate rats. Maximum uterine growth response produced by the grass juice concentrate indicated that 1 cc contained approximately the equivalent of 0.016 mg. of estradiol benzoate.

RUMEN MICROORGANISM GROWTH REGULATION FACTOR(S) FOUND IN EXTRACTS OF RUMEN CONTENTS AND OF SOME RUMINANT FEEDS. G. B. Garner, M. E. Muhrer and W. H. Pfander, *University of Missouri.*

Extracts of rumen fluid and fermented feeds contain a factor(s) which stimulates cellulose digestion, *in vitro,* and at the same concentration is effective in inhibiting the growth of undesirable microorganisms sometimes found in the rumen. This factor(s) has been found in bovine and ovine rumen fluids, in feeds fermented by rumen microorganisms and in some types of silage. The effect of the factor(s) on cellulose utilization was determined *in vitro* using washed rumen microorganisms and a basal medium consisting of cellulose, urea, casein, ammonium carbonate, glucose, cobalt chloride and artificial saliva. In a 24 hour period, cellulose digestion was increased 25 to 30% over the basal. By standard turbidimetric methods the extracts were found to be effective in inhibiting the growth of *Pseudomonas aeruginosa* and *Aerobacter aerogenes.* These organisms were previously isolated from animals with rumen dysfunction. However, under the same conditions, the growth response of washed rumen microorganisms was rapid and logarithmic in relation to dosage when the organisms were obtained from healthy animals. These studies indicate that there is at least one factor present in rumen fluid and in fermented feeds that inhibits the growth of microorganisms which sometimes contaminate the rumen as well as enhances the growth and function of desirable rumen microorganisms.

COB PORTION OF GROUND EAR CORN AS SOLE ROUGHAGE FOR FATTENING CATTLE. H. B. Geurin, J. C. Thompson, H. L. Wilcke and R. M. Bethke, *Ralston Purina Company.*

A series of cattle feeding tests have evolved a new concept for effective utilization of the roughage of cobs in ground ear corn conducive to maximum gains and finish. Several previous tests with 275 steers have shown that cattle on ground ear corn plus hay gained less (1.99 vs. 2.11 lb. per day) and carried less finish than comparable steers on shelled corn. In three later tests 46 steers received a complete ration of corn, a Vitamin A and trace mineral fortified supplement, and a minimum of ground cobs. Their daily gains were 2.24, 2.45, and 2.72 lb. In a following test 30 yearling steers received either shelled corn, hay and supplement; ground ear corn and supplement; or ground ear corn and the above fortified supplement. The respective daily gains and selling prices were 2.55, 1.65, 2.53; $26.25, $24.00, and $26.25. This year 17 steer calves received either shelled corn, alfalfa hay and supplement; or ground ear corn, alfalfa hay and supplement with the hay removed and the above fortified supplement fed during the finishing period. The respective daily gains and increases in market grade during the starting period; the same data for the finishing period; the shipping shrink; the selling prices; the USDA carcass grades and the chilled carcass dressing percent were as follows: 2.27, 2.29; 2 1/3, 1 2/3; 2.03, 2.07; 1 2/3, 1 2/3 +; 2.99, 2.92; $23.75, $23.75; Prime, Low Prime; 63.0 and 62.0. The cob portion of ground ear corn was effective as the sole roughage.

ILOTYCIN AS A FEED INGREDIENT FOR GROWING-FATTENING SWINE. L. Gillespie and H. D. Wallace, *Florida Agricultural Experiment Station.*

Fifty weanling pigs (25 Hampshires, 25 Durocs) were divided into five experimental groups. Each group was subdivided into two pens, one containing five Hampshires and one containing five Durocs. A basal ration consisting of 77 parts corn, 21 parts soybean oilmeal, 1 part limestone, 0.5 parts steamed bonemeal, and 0.53 parts salttrace mineral was used. It was fortified with thiamin, riboflavin, niacin, pantothenic acid, pyridoxine, choline, folic acid and B_{12}. The pigs were self-fed in concrete pens which were washed daily. The experimental treatments were as follows: Group I—Basal; Group II—Basal + 2 mg. Ilotycin per pound; Group III—Basal + 6 mg. Ilotycin per pound; Group IV—Basal + 10 mg. Ilotycin per pound; Group V—Basal + 20 mg. Ilotycin per pound. At the end of 8 weeks average daily gains and feed conversion figures for Groups 1 through 5 respectively were: 1.28, 306.5; 1.29, 289.9; 1.30, 296.4; 1.30, 290.7; and 1.28, 292.0.

EFFECTS OF DIETARY FLUORINE ON FLUORINE CONTENT OF BONES IN CATTLE AND SHEEP. J. M. Griffith, C. S. Hobbs, R. P. Moorman, G. M. Merriman, R. P. Gregory, Jr. and W. H. MacIntire, *University of Tennessee.*

The fluorine content of ribs from 59 cows, that were fed various levels of fluorine in the ration varying from control ration (8 ppm) to ones containing as much as 100 ppm, showed a direct relationship to the level of fluorine fed. Fluorine content of the ribs, based on rib biopsies, at the end of 26 months on feed showed little or no difference from those of the same animal at the end of 62 months on experiment. These data with other data may indicate that there is a different saturation point for the different levels of fluorine ingested. The fluorine content of bones from 5–6 month old calves, that had nursed these cows, showed no significant differences that could be attributed to the fluorine intake of the dams. This indicates that there is no significant transfer of fluorine through the placenta

or through the milk. However, the fluorine content of bones from calves running with their dams on pasture was directly related to the flourine content of the pasture which was probably due to the calf consuming forage containing fluorine. A comparison of the fluorine content of the bones of cows, ewes and rats fed various levels of fluorine with and without the addition of aluminum sulfate, shows that aluminum sulfate fed as an alleviator significantly lowered the amount of fluorine stored in the bones and decreased the effect of fluorine on developing teeth. A comparison of the fluorine content of the rib, jaw and leg bones from cattle receiving increased levels of fluorine continuously (100 ppm to 1200 ppm), in general, showed the rib to be the highest, the jaw somewhat lower and the leg the lowest.

RESPONSES OF LAMBS FED VARIED LEVELS OF DIETHYLSTILBESTROL. W. H. Hale, C. D. Story, C. C. Culbertson and Wise Burroughs, *Iowa State College.*

Six individual feeding experiments have been completed in which diethylstilbestrol was included in the rations of fattening lambs at levels from 0.5 γ to 1200 γ per lb. of ration. The lambs were fed a mixed ration containing corn, soybean oil meal, molasses and alfalfa. In five of the six trials a response in weight gain was obtained at or near the 2.0 γ level. For all six trials this response was 19% over the controls. At levels between 3.0 γ and 600 γ no consistent response was found but quite often a depressing action was noted. The 600 γ and 1200 γ levels increased weight gains 20 and 28% respectively over the controls. However the above results were not verified in 2 group feeding trials. Diethylstilbestrol administered orally did not appear to have an appreciable effect upon carcass quality. Measurement of certain secondary sex organs of the wethers and uterine measurement on a limited number of ewe lambs showed that the orally administered diethylstilbestrol exerted a physiological activity. Data collected suggest that for optimum performance the oral dose level may be affected by seasonal difference.

CHEMICAL AND ENZYMATIC PREPARATIONS OF PROTEIN HYDROLYSATES STIMULATORY TO CELLULOSE DIGESTION BY RUMEN MICROORGANISMS. Glen Hall, Edmund W. Cheng, William H. Hale and Wise Burroughs, *Iowa State College.*

Unidentified factors present in ruminant feeds stimulatory to rumen microorganisms in the utilization of fibrous feeds by cattle and sheep have been noted by numerous investigators. These unidentified factors have not been isolated nor chemically characterized. During the course of studies of this nature it was found that these factors were water soluble and often occurred in largest quantities in feeds either rich in protein or in natural feeds rich in non-protein-nitrogen. Attempts were made in this laboratory to alter the structure of water insoluble proteins in such a way that these unidentified stimulatory factors could be produced at will by such alterations. Purified casein which is not water soluble and which in the unaltered state exhibits no stimulatory properties was accordingly subjected to a mild hydrolysis either by acid, alkali, or enzymatic treatment. In each instance mildly hydrolyzed casein exhibited a highly favorable influence upon a washed suspension of rumen microorganisms in the digestion of cellulose. Severe hydrolysis of casein, breaking down to the free-amino acid state, resulted in complete loss of such activity. This suggests that the active principles produced and probably the unidentified factors present in certain cattle feeds are water soluble peptides. Other isolated proteins and protein rich materials such

as soybean protein, chicken feathers, hair, and blood meal which showed no stimulatory properties before hydrolysis were found to be active when hydrolyzed similar to hydrolysis performed on casein.

THE UTILIZATION BY CATTLE OF CALCIUM FROM VARIOUS SOURCES. Sam L. Hansard, H. M. Crowder and C. L. Comar, *University of Tennessee.*

Utilization efficiency of calcium from various organic and inorganic sources has been measured in young and mature cattle from conventional balance data corrected for endogenous losses. Results of radio-chemical procedures with paired Hereford calves and paired mature steers, in 16 balance trials on natural farm rations, indicated fecal endogenous calcium excretion to average 15 ± 2 and 16 ± 3 mg. of calcium per kilogram body weight, respectively. After 30 days on a low calcium ration of orchard grass hay and corn, endogenous fecal losses were reduced to 10 ± 2 and 12 ± 3 mg. per kilogram body weight for calves and steers respectively. Results from the incorporation of various sources of calcium into basal grain rations to give normal dietary calcium levels for the two age groups indicated that there was more difference in utilization due to age than to source of calcium. Absorption values for monocalcium phosphate, bone meal, dicalcium phosphate, calcium chloride, calcium carbonate and limestone (five) in forty-four trials ranged from 45 to 65 and 36 to 57% for young and mature animals respectively. When good grades of orchard grass, alfalfa or lespedeza hay were used as the dietary source of calcium a similar utilization value was obtained. However, good alfalfa hay fed to mature steers at optimum calcium levels resulted in a decrease in utilization, emphasizing the necessity for availability measurements at dietary levels closely approximating calcium requirements.

EFFECTS OF FEEDING VARIOUS LEVELS OF FLUORINE, DEFLUORINATED PHOSPHATE AND GRAIN TO DAIRY HEIFERS. Lorin E. Harris, George E. Stoddard, D. A. Greenwood, J. L. Shupe, George Q. Bateman and Wayne Binns, *Utah Agricultural Experiment Station.*

Thirty-two Holstein heifers weighing initially 180 to 274 lb. were assigned to treatments consisting of four levels of fluorine (basal diet, 25, 50, and 100 ppm based on hay intake), two levels of calcium and phosphorus (1% and 3% defluorinated phosphate in the grain mixture), and two levels of grain (two and four pounds daily until calving then grain at the rate of 5.2 lb. for each lb. of butterfat, and 7 lb. for each lb. of butterfat). At the end of 28 twenty-eight-day periods the results were as follows: The heifers consuming the larger amounts of grain gained more, were higher at the withers, and consumed more dry matter during the early stages, but there were no differences after 784 days. The heifers on less grain consumed more hay. Balance trials conducted 68 to 113 days after experiment started showed that as the fluorine in the diet increased the amount of fluorine stored in the body increased. Heifers on 4 lb. of grain stored less fluorine than those on 2 lb. of grain. Three percent defluorinated phosphate in the diet promoted more storage of fluorine than one percent. Heifers on 2 lb. of grain digested more protein and crude fiber and stored more nitrogen than those on 4 lb. Heifers on 4 lb. of grain digested more ether extract and the diet contained more total digestible nutrients than those on 2 lb. Heifers on one percent defluorinated phosphate digested more dry matter than those on three percent. Treatments did not affect nitrogen storage, phosphorus balance, or digestion of cellulose, lignin, or energy. Balance trials were also conducted 469 to 509 days after the experiment started. The results were similar. Animals on the higher levels of fluorine have shown some tooth and bone changes.

EFFECT OF SUPPLEMENTAL MN IN THE DIET OF LAMBS ON HB CONCENTRATION. R. H. Hartman, Gennard Matrone and G. H. Wise, *North Carolina Agricultural Experiment Station.*

Studies of the effects of different levels of manganese on the nutrition of the lamb revealed that excessive dietary manganese had a depressing effect on concentrations of hemoglobin and of serum iron. This response was demonstrated in two experiments. In the first the basal diet consisted of whole milk fortified with vitamins and minerals. When this diet was supplemented with as little as 45 p.p.m. of Mn, hemoglobin and serum iron levels were decreased. Higher levels of manganese supplementation (up to 5000 p.p.m.) were associated with increased concentrations of copper and decreased concentrations of iron in the livers, spleens and kidneys. In the second experiment the basal diet consisted chiefly of 60% hay, 35% corn sugar and 5% refined cottonseed oil (Wesson). The hemoglobin concentration of the experimental lambs was decreased to 5 or 6 gm./100 ml. of blood by bleeding. The lambs thus rendered anemic were randomized to diets containing 0, 1000 and 2000 p.p.m. of manganese. Hemoglobin regeneration was markedly retarded and serum iron levels depressed in lambs fed the diets containing either 1000 or 2000 p.p.m. manganese.

A STUDY OF THE EFFECT OF AGE ON THE PROTEIN AND SULFUR-CONTAINING AMINO ACID REQUIREMENTS OF THE RAT. E. W. Hartsook and H. H. Mitchell, *University of Illinois.*

Thirty weanling male albino rats were fed ad libitum a 20% protein ($N \times 6.25$) diet, adequate in all known nutrients, from weaning to an age of 102 days from birth. The protein was casein supplemented with 3.2% DL-methionine. Body weight and food consumption of each animal were recorded weekly and a quadratic equation was fitted to the regression of food consumption on body weight. The two rats nearest the group mean weight were killed at intervals of weight increase of 20–25 gm. and carcass nitrogen, methionine, and cystine contents were determined. Equations of the type $C = a - be^{-ct}$, where C is total amount of carcass component, t is age in days, e is the base of natural logarithms, and a, b, and c are constants, were fitted to the data. Evaluation of first derivatives of these equations furnished accurate estimates of the daily rate of carcass deposition of the nutrients of interest at any age t. To these quantities were added careful estimates of daily maintenance requirements and of daily amounts of nutrients lost as shed hair, yielding daily net requirements. Dietary requirements were calculated by using an assumed BV of 90. The dietary protein and methionine + cystine requirements decreased exponentially as the animals increased in age from 30 to 85 days. The dietary requirements at ages of 30 and 85 days were 27.8 and 6.8%, respectively. The dietary methionine + cystine requirements at the same ages were 1.28 and .30%, respectively.

THE UTILIZATION OF NON-PROTEIN NITROGEN BY GROWING-FATTENING PIGS. V. W. Hays, D. V. Catron, G. C. Ashton, V. C. Speer and C. H. Liu, *Iowa State College.*

The purposes of the experiment were to determine the levels of NPN (urea) a pig can tolerate and if a portion of the pre-formed protein can be replaced by NPN. The tolerance level, toxicity effects and utilization of urea were studied. The experiment involved 48 pigs individually fed in wire floored crates. Poland China x Landrace x Duroc cross pigs were randomly allotted by outcome groups based on weight and sex into a 2×6 factorial experiment. Average initial weight was 23 lb. and the experiment terminated as each pig individually reached 75 lb. The

ration treatments were based on a 16% protein corn-soybean oil meal ration in which 0, 2.5, 5, 10, 20 and 40% of the protein equivalent came from urea. Each level was fed with and without chlortetracycline to 4 animals per ration treatment or 8 animals per level of urea. At the termination of the experiment, blood samples were taken and analyzed for ammonia and urea; feces and urine were collected and nitrogen balance determinations were made. The animals on the 0, 10 and 20% urea levels were slaughtered and kidneys were inspected for possible renal damage. The results show that the pig can tolerate up to 10% of the protein equivalent as urea without seriously affecting growth rate or feed efficiency. On the lower levels of urea, antibiotics increased rate of gain and feed efficiency; however, on the higher levels (20 and 40% protein equivalent as urea) the antibiotics seemed to decrease the urea tolerance and/or utilization.

THE CALCIUM AND PHOSPHORUS COMPOSITION OF THE ANIMAL BODY AS INFLUENCED BY THE CHEMICAL COMPOSITION OF PLANTS GROWN ON SOILS OF VARYING PHOSPHORUS LEVELS. W. W. Heinemann, M. E. Ensminger, W. E. Ham and J. E. Oldfield, *State College of Washington.*

Domestic rabbits were used to determine effects of alfalfa hay produced on soil with (1) low available phosphorus and (2) high available phosphorus on (a) efficiency of reproduction, (b) growth rate, (c) inorganic phosphorus content of blood serum, (d) breaking strength and calcification of skeletons, and (e) calcium, phosphorus, sodium, magnesium, fat, and total ash of bones. Alfalfa hays containing 0.12% and 0.26% phosphorus were used as 86% of the rations. The following differences were observed on the low phosphorus hay: (1) significantly retarded growth, (2) lower mature body weights, (3) impaired breeding efficiency, (4) significantly higher levels of calcium, phosphorus, fat, and ash in the skeletons of ten-week-old animals and significantly lower levels of ash in skeletons of mature animals, (5) extremely low breaking strength of the humerus, approaching 100% difference in the third and fourth generations, and (6) abnormal bone structure and ossification as revealed by X-ray photographs. There was no change in inorganic phosphorus in the serum. It is concluded that where the phosphorus content of alfalfa hay produced under Columbia Basin conditions is of the order of 0.12% it is not adequate for normal animal nutrition. This condition can be corrected by proper phosphate fertilization of the soil. Results of this study establish that there is a positive soil-plant-animal relationship among (a) the soil on which alfalfa hay was produced for this work, (b) the forage crop produced thereon, and (c) the animal organism.

THE DISTRIBUTION OF AUREOMYCIN IN THE BODY OF DAIRY CALVES SHOWING A GROWTH RESPONSE WHEN THE ANTIBIOTIC IS ADMINISTERED ORALLY OR INTRAMUSCULARLY. H. H. Hester, Jr., F. T. Landagora and L. L. Rusoff, *Louisiana Agricultural Experiment Station.*

An investigation was made of the absorption, distribution, and excretion of aureomycin in the body of calves administered nutritional levels of the antibiotic and showing a growth increase of 19% above the controls at 16 weeks of age. Three comparable calves from each of the three groups (Group 1—Control; Group 2—Oral Aureomycin 50–90 mg. daily; Group 3—Injected Aureomycin, 400 mg. weekly) were slaughtered and samples taken of body fluids, organs, tissues, and contents of the gastro-intestinal tract for aureomycin assay. The antibiotic was present in the blood plasma, bile, and urine of the aureomycin administered calves. Highest concentrations were found in the urine of injected calves and the lowest levels in the blood plasma. The livers and kidneys of the injected calves

contained measureable amounts of aureomycin while a trace was present in the liver and kidney of one calf of the oral-fed group. The spleen, thymus, pituitary, and muscle showed no detectable antibiotic in any of the calves. No aureomycin was found in the rumen of the injected calves. This finding is in opposition to the theory that explains the mode of action of aureomycin thru alteration of the rumen microflora. A definite pattern of distribution was established in the intestinal tract of the calf. The antibiotic increased in concentration from the upper portion to the lower portion of the small intestine, and decreased in concentration between the lower small intestine and the anal end of the large intestine.

THE INFLUENCE OF SEVERAL ANTIBIOTICS ON THE PERFORMANCE OF YOUNG DAIRY CALVES. D. E. Hogue, R. G. Warner, C. H. Grippin and J. K. Loosli, *Cornell University*.

The influence of aureomycin, streptomycin and a mixture of bacitracin and penicillin (4:1) on young dairy calves was measured using 96 grade Holstein heifer calves in two replicates of a $2 \times 10 \times 2$ factorial experiment. Additional variables included: two milk levels (350# or 175#), three antibiotic levels (10, 20, or 40 mg./100 lb. body weight/day) and two durations of antibiotic feeding (7 or 16 weeks). The data were evaluated at the end of 7 and 16 weeks. All antibiotics significantly ($p < 0.05$) increased the daily gain (lb./day), growth of heart girth (inches) and feed efficiency (lb. TDN/lb. gain) and decreased the days abnormal feces for the first 7 weeks. The mean values for each were respectively: controls, 0.78, 3.8, 2.34, 4.8; aureomycin, 1.00, 4.7, 1.91, 2.3; bacitracin-penicillin, 0.92, 4.5, 2.06, 2.1; streptomycin, 0.92, 4.7, 2.06, 2.2. At 16 weeks there were no significant differences between treatments for the observations mentioned above. The bacitracin-penicillin group consumed significantly less starter than the others ($p < 0.05$). The 350# milk group gained 0.98 and 1.17 lb./day during the 7 and 16 week periods respectively. The 175# group gained 0.83 and 1.11 lb/day for the same periods. Differences between milk levels were significant ($p < 0.05$). Both aureomycin and streptomycin appeared to compensate for a reduced milk intake while having little influence on nonscouring calves receiving 350# of milk. These experiments showed no advantage for any particular level of antibiotic nor for their feeding past 7 weeks of age.

ACUTE NICOTINIC ACID DEFICIENCY IN THE CALF. John H. Hopper and B. Connor Johnson, *University of Illinois*.

A nicotinic acid deficiency was produced and studied in young ruminants by *ad libitum* feeding one-day-old calves a synthetic milk diet in which acid-hydrolyzed casein plus sufficient "vitamin-free" casein to provide 0.17% tryptophane was used as a source of protein. Calves receiving this diet plus all the vitamins continued to grow and eat well, whereas the calves deprived of nicotinic acid exhibited very marked symptoms by the second day; namely, severe scouring, dehydration, and loss of appetite. A rapid recovery from this condition was observed following oral administration of nicotinic acid.

AMMONIATED CITRUS PULP IN THE FATTENING RATION. W. G. Kirk, George K. Davis and Fentress M. Peacock, *Florida Range Cattle Experiment Station*.

Ammoniated citrus pulp is a new product. To determine its value as a source of equivalent protein, three dry lot trials with steers have been completed. The protein equivalent of ammoniated pulp on a D.M. basis was 12.11, 15.12 and 13.87% for Trials 1, 2 and 3 respectively, while plain pulp contained 7.06%

protein. Steers were fed Pangola hay and 2 lb. citrus molasses daily in all trials. Trial 1: Yearling steers fed a ration of 3 parts ammoniated pulp and 1 part cottonseed meal gained 2.13 lb. daily in a 49-day period while in the next 56 days daily gains were 1.45 lb. Replacing 44 parts of the original ration with a mixture containing plain pulp, cottonseed meal, cracked corn and alfalfa meal increased daily gains to 2.14 lb. in the next 31 days while steers fed the check ration gained 2.02 lb. daily. Trial 2: Two-year-old steers fed 80 parts ammoniated pulp and 20 parts cottonseed meal gained 1.81 lb. daily in 87 days, while those fed the check ration gained 2.61 lb. Replacing 17 parts of the ration with corn meal increased the daily gains of steers fed ammoniated pulp to 2.90 lb. in a 31-day period. Trial 3: Steer calves fed 77 parts ammoniated pulp, 18 parts cottonseed meal and 5 parts alfalfa meal made a daily gain of .80 lb. in the first 28 days while those fed the check ration gained 1.65 lb. In the next 112 days 15% of corn meal was used in both rations, increasing daily gains of steers fed ammoniated pulp and plain pulp to 2.00 lb. and 2.32 lb. respectively.

RELATIONSHIPS BETWEEN LEVEL OF PROTEIN, MOLASSES AND QUALITY OF HAY IN RATIONS FOR FATTENING CATTLE. Earle W. Klosterman, Orville G. Bentley, A. L. Moxon and L. E. Kunkle, *Ohio Agricultural Experiment Station.*

Twelve lots of seven steer calves each were fattened in dry lot on ground ear corn and minerals including trace mineralized salt. Six of these lots were fed good quality mixed clover and timothy hay and six lots late cut timothy hay. Three levels of protein, with and without one pound of molasses per head daily, were fed to the steers which received each type of hay. There were highly significant differences in average daily gains between lots of steers fed 1.5, 0.75 lb. and no soybean oil meal per head daily. There were also highly significant differences in rates of gain between steers fed the good mixed hay and those fed the poor timothy hay. The interaction between quality of hay and amount of soybean oil meal fed was highly significant—the differences between the two types of hay increased as the amount of soybean oil meal fed was decreased. Molasses was of most benefit when fed with 0.75 lb. soybean oil meal, however, there was no significant improvement in rate of gain as a result of feeding molasses nor were the interactions between molasses and level of protein or molasses and quality of hay significant. Feed costs averaged lowest for steers fed good quality hay without molasses.

UPTAKE OF RADIOSULFUR BY SHEEP GIVEN REPEATED DOSES OF LABELED SODIUM SULFATE. Roman Kulwich, Lee Struglia, Henry C. Bourne and P. B. Pearson, *U. S. Department of Agriculture.*

The two lambs used in this study were three months old at the start of the experimental period. They were each fed pelleted rations ad libitum. Lamb A, a wether, received a ration with soybean protein as the chief source of protein while lamb B, a ewe, received a similar ration in which urea, at a level of 1.2%, replaced part of the protein. A total of 11 doses of radiosulfur in the form of sodium sulfate were administered in gelatin capsules with a balling gun to each lamb over a period of 24 days. Each dose contained 0.7 milligrams of carrier sulfate and approximately 0.6 millicurie of radiosulfur. These animals were sacrificed 7 days after receiving the last dose. The excreta collected during the 7 days following the final dose were analyzed for radiosulfur content and it was found that in the case of lamb A the urine contained radiosulfur equivalent to 72% of a single dose while the feces radiosulfur content was equivalent to 39% of a single

dose. The corresponding values for lamb B were 55% and 59%, respectively. Radioautographs which were made of wool fibers showed that approximately a half inch length of the fiber, extending upward from the base, contained radiosulfur. The concentration of radiosulfur in wool was higher than that found in any of the tissues analyzed. For most of the tissues studied, lamb B had higher tissue concentration of radiosulfur than lamb A.

EFFECT OF TYPE AND AMOUNT OF DIETARY LIPIDS AND PROTEIN ON GROWTH AND INCIDENCE OF DIARRHEA OF CALVES FED SEMI-SYNTHETIC DIETS. M. R. Lambert, R. S. Allen, N. L. Jacobson and Ruth M. Ward, *Iowa State College.*

To study further the lipid requirements of young dairy calves, a basic semi-synthetic diet containing lactose, salts, vitamins, crystalline aureomycin HCl, and soybean protein (Drackett) plus either vitamin-test casein or 0.2% methionine was employed. A lipid-free liquid diet containing 9% lactose and 6.25% Drackett (plus methionine) fed at the rate of 10% body weight daily produced essentially no weight change over an 8-week period. Incidence of diarrhea was low during the initial week but increased thereafter. At about 7 weeks of age the calves exhibited other fat deficiency symptoms (rough hair coat, partial loss of hair, emaciation). At this stage, recovery after introduction of lipids into the diet was slow and sporadic, although in most instances a marked reduction in diarrhea occurred. When 1.8% butter oil and 0.2% lecithin were added to the semi-synthetic diet from the beginning of the experimental period, incidence of diarrhea was very low (even though there was no reduction in non-lipid dietary components) and no fat deficiency symptoms were exhibited. Lecithin alone (approx. 0.2%), although ineffective either in preventing or in promoting recovery from the lipid-deficiency syndrome, resulted in blood plasma fat (Allen) values of about the same magnitude as those resulting from the feeding of 2.0% butter oil. A diet containing 1.75% Drackett protein plus 1.75% vitamin-test casein supported better growth than one containing 3.5% Drackett protein plus 0.2% methionine.

THE EFFECT OF VARIOUS SURFACTANTS AND AUREOMYCIN ON THE DIGESTIBILITY OF FEED NUTRIENTS BY DAIRY STEERS. C. A. Lassiter, T. W. Denton, C. L. Davis and J. A. Taylor, *University of Kentucky.*

Four yearling dairy steers (650–800 lb.) were used in a standard digestion trial to study the effect of various surfactants and crystalline aureomycin on the digestion of feed nutrients. A ten-day preliminary period and seven-day collection period were used. An alfalfa hay and a simple concentrate mixture were fed, each one supplying one-half of the dry matter. The experimental products fed, order of feeding, and amount of supplement fed are as follows: (1) basal, (2) Ethomid C/15, 0.1% of dry matter fed; (3) Arquad HT, 0.0125% of D.M.; (4) aureomycin, 0.5 gm./day; (5) Ethomid C/15, 0.1% of D.M., plus aureomycin, 0.5 gm./day. None of the surfactants or aureomycin appeared to have any effect on the appetite, consistency of feces or over-all condition of the steers. The supplements did not have any significant effect upon the digestibility of dry matter, crude protein, either extract or nitrogen-free extract. The average coefficients for the digestibility of dry matter were: basal, 64.0%, Ethomid C/15, 61.6%, Arquad HT, 66.9%, aureomycin, 60.5%, and Ethomid C/15 plus aureomycin, 63.9%. The coefficients of apparent digestibility for crude fiber in the same order were: 35.5, 35.8, 41.1, 22.7 and 37.5%. Aureomycin reduced the digestibility of crude fiber by an amount that was significant at the 5% level of probability.

THE EVALUATION OF AN UNIDENTIFIED GROWTH FACTOR AND TERRAMYCIN FOR YOUNG DAIRY CALVES. C. A. Lassiter, T. W. Denton, J. W. Rust and D. M. Seath, *University of Kentucky.*

Thirty-two 2-day old calves (8 Jerseys and 24 Holsteins) were used in studies to evaluate a source of an unidentified growth factor for young dairy calves. This factor has been shown by other workers to promote the growth rate of chickens and swine. The source of the unidentified growth factor was a mixture of products from terramycin and streptomycin fermentations. Crystalline terramycin and a combination of terramycin and the unidentified growth factor were also included in the studies. The calves were raised to 86 days of age on a limited milk-starter-hay system. Milk was fed at the rate of 8% of body weight for the first 21 days and gradually reduced until the calves were off milk at 49 days of age. All supplements were fed in the milk for the first 49 days and in the starter thereafter. The unidentified factor was fed at the level of 2½% of the starter except in a few cases when it was fed at the 1.25% level. Terramycin was fed at the rate of 75 mg. per day for the first 49 days and 24 mg./lb. of starter thereafter. Average daily gains, feed efficiencies, (lb. feed/lb. gain) and scouring data (days per calf) of the calves on the various treatments were as follows: control—1.20, 2.57, 1.25; terramycin—1.15, 2.59, 0.75; unidentified growth factor—1.18, 2.64, 2.00; terramycin plus unidentified growth factor—1.27, 2.56, 1.12. The presence of the unidentified growth factor in the respective rations tended to scour the calves for the first three weeks if fed at the 2½% level. This condition was eliminated by reducing the amount of the factor fed to a 1.25% level for the first three weeks. None of the differences in daily rate of gain or efficiency of feed utilization was statistically significant.

PROTEIN LEVELS FOR PIGS AS STUDIED BY GROWTH, SELF-SELECTION AND NITROGEN RETENTION. J. W. Lassiter, S. W. Terrill, D. E. Becker and H. W. Norton, *University of Illinois.*

A protein supplement consisting of soybean oil meal, meat and bone scrap, menhaden fish meal and condensed menhaden fish solubles, fortified with minerals, vitamins and antibiotics, was used with yellow corn to test protein levels of 10, 12, 14 and 16% from weaning to 200 lb. on 40 pigs group-fed *ad libitum* in drylot. In pasture tests a protein supplement of soybean oil meal, meat and bone scrap and menhaden fish meal was used to test levels of 8, 10, 12 and 14% on 40 pigs. In drylot the gains were significantly slower on 10% protein than the higher levels, where the gains increased as the protein level increased (not statistically significant). On pasture, the average gains of pigs on the 14% level was below the value extrapolated from the lower levels, suggesting that the requirement is between 12 and 14%. Using the same two protein supplements, respectively, in free-choice feeding 18 pigs in drylot selected an average of 14.5% protein from weaning to 100 lb. and 11.8% from 100 to 200 lb. on pasture 10.8 and 9.9%.

BLOOD GLUCOSE AND ACETONE BODIES, LIVER GLYCOGEN AND FAT OF NORMAL, FASTED AND KETOTIC COWS. E. C. Leffel and J. C. Shaw, *University of Maryland.*

Twenty-eight cows were fed on high, medium or low protein rations for 60 days before parturition. After parturition, cows were either full fed or fasted. Blood glucose and acetone body levels were determined. Liver samples were obtained by biopsy and analyzed for glycogen and fat. Before parturition, average blood glucose was 47 mg. and blood acetone bodies 2.7 mg. per 100 ml. of blood, average liver glycogen content was 2.8% and liver fat 3.8%. Fasting for 10 days immediately

postpartum resulted in a marked hypoglycemia and ketonemia (blood glucose 22 mg. %, acetone bodies 23 mg. %), while cows liberally fed during the same period failed to show a significant hypoglycemia or ketonemia (blood glucose 41 mg. %, acetone bodies 6.3 mg. %). Fasting also resulted in a much lower liver glycogen and higher liver fat (0.25% and 15.2%) when compared to more liberal feeding (glycogen 2.3%, fat 6.5%). The level of protein fed did not affect the blood and liver substances in this study. Fasted cows did not show symptoms typical of ketosis. Cows with clinical ketosis exhibited low levels of blood glucose and liver glycogen and high levels of blood acetone bodies and liver fat, though more variation was observed in these field cases than was found in fasted cows.

THE APPARENT DIGESTIBILITY OF THE CRUDE PROTEIN OF THE PIG RATION AS A FUNCTION OF ITS PROXIMATE COMPOSITION. L. E. Lloyd and E. W. Crampton, *Macdonald College* (*McGill University*).

Rat feeding studies on the effect of proximate composition on the apparent digestibility of energy yielding components of the diet have been extended to include experiments with swine. Whereas in the case of rats the apparent digestibility of protein was almost completely a function of protein level of the diet, the data for swine indicate that the crude fibre inherent in the feedstuffs appreciably influences the fecal output of nitrogen. This species difference might be attributed to the fact that a purified crude fibre, methyl cellulose, was added to the rat diet, while the fibre of the swine rations occurred naturally in the feeds used.

ANTIBIOTICS IN RUMINANTS. BACTERIOLOGY—ARTIFICIAL RUMEN—GROWTH STUDIES. H. G. Luther, C. R. Adams, W. B. Hardie, Z. Zollie, W. M. Reynolds and H. E. Downing, *Chas. Pfizer & Co., Inc.*

Feeding and artificial rumen trials have been conducted to determine the effect of graded levels of oxytetracycline (Terramycin) on both roughage and concentrate type rations for cattle. Evaluation of antibiotic level on rumen flora and its function were judged by microbiological observation, culture methods, artificial rumen methods and growth studies. The levels of antibiotics used ranged from 50 ppm in the feed to 5 gm. daily dosage administered by capsule. Microbiological observations and culture methods revealed an alteration of rumen flora with antibiotic administration. The change was greatest in the early period but gradually returned to its original state as the trial progressed. Similar pattern of cellulolytic activity was evident with both glass type and permeable sac miniature artificial rumen methods. The animals on a high concentrate ration showed a return to normal activity approximately 48 hours after consuming a ration containing 75 to 150 mg. of Terramycin per day. The animals on a high roughage ration showed a return to normal activity after approximately 72 hours on a ration containing the same levels of Terramycin per day. Parallel animal growth studies reveal the addition of these same levels in the ration resulted in improvement in daily rate of gain and efficiency of feed utilization.

PALATABILITY STUDIES ON CREEP FEED FORMULATIONS FOR SUCKLING PIGS. F. A. McMillan and H. D. Wallace, *Florida Agricultural Experiment Station.*

The palatability of eight creep feed mixtures was studied using a total of 217 suckling pigs. Mixture 1 was composed of 46% ground yellow corn, 30% ground rolled oats, 18% soybean oilmeal, 5% dried skimmilk and appropriate vitamin-mineral-antibiotic fortifications. In mixture 2 the ground yellow corn was replaced by partially dextrinized, partially gelatinized ground yellow corn and in Mixture 3

similarly treated corn with the germ and bran removed was used. Mixtures 4 through 8 were similar to mixture 1 except that special ingredients replaced an equal weight of ground yellow corn. Mixtures 4 and 5 contained 10% of prime stabilized beef tallow; mixture 6, 10% of citrus molasses; mixture 7, 10% of black strap molasses; and mixture 8, 10% of cane sugar. All mixtures were fed in the pelleted form except 5. The sows and litters were placed on pasture when the pigs were about one week of age and fed in groups of 1–5 sows and litters per lot. The eight feed mixtures were offered free choice from small self feeders in covered portable pig creeps. Thirty-eight of the pigs were weaned at 6 weeks of age and the remaining 179 at 8 weeks. The comparative consumption for the various mixtures was: 1—.90%; 2—.67%; 3—2.32%; 4—33.42%; 5—11.84%; 6—.52%; 7—1.59%; and 8—48.73%.

THE THIAMINE REQUIREMENT OF THE BABY PIG. E. R. Miller, D. A. Schmidt, J. A. Hoefer and R. W. Luecke, *Michigan State College.*

A total of 40 animals was used in two feeding trials to determine the thiamine requirement of the baby pig. The pigs were taken from the sow at three days of age and following a depletion-adjustment period on a thiamine-free, synthetic milk diet, were placed on diets supplemented with five different levels of thiamine, ranging from the basal no thiamine level to 2.0 mg. per kilogram of solids. The feeding method was essentially *ad libitum*. Data on individual growth response and dietary intake were recorded. Analysis of the data indicates that the dietary thiamine requirement of the baby pig for optimum growth is 1.0 to 1.5 mg. per kilogram of solids intake. Good gaining ability was rapidly restored to deficient animals which received thiamine injections for a period. However, evidence of irreversible changes in heart tissue was still present upon post mortem examination.

THE USE OF GROUND CHICKEN LITTER AS A NITROGEN SOURCE FOR FATTENING STEERS AND GESTATING-LACTATING EWES. P. R. Noland, B. F. Ford and M. L. Ray, *University of Arkansas.*

Two groups of twelve gestating-lactating ewes were fed either soybean meal or ground chicken litter (30% protein equivalent) as sources of supplementary protein for a 156 day period. No difference existed in the average final weight of the two groups of ewes. Average daily gain in the lambs from birth to termination of the trial was 0.44 $\pm$ .008 lb. for the soybean meal fed group and 0.42 $\pm$ .010 lb. for the chicken litter fed group. No deleterious effects were observed in either of the two groups of ewes or lambs. Non-protein nitrogen content of the blood from the ewes 24 hours post-feeding was 25.8 $\pm$ 6.7 mg. per 100 cc. in the soybean meal group and 25.1 $\pm$ 3.4 in the chicken litter group. The three hours post-feeding content was 33.3 $\pm$ 7.7 and 26.9 $\pm$ 1.2 respectively. Twenty fattening steers were pair-fed for equal feed and equal nitrogen intake using cottonseed meal or ground chicken litter as the supplementary sources of nitrogen. During a 56 day trial, steers fed the cottonseed meal-grain mix gained 2.14 lb. per day and those fed the chicken litter-grain mix gained 1.78 lb. daily.

THE RELATIONSHIP OF UNIDENTIFIED FACTORS FOUND IN FISH LIVER, FISH SOLUBLES, AND A FERMENTATION PRODUCT WHEN ADDED TO AN ALL-VEGETABLE RATION FOR GROWING AND FATTENING SWINE. P. R. Noland, E. L. Stephenson and J. J. Miner, *University of Arkansas.*

A growth response was obtained from the addition of a fermentation product to a corn-soybean meal type ration fed to weanling pigs in drylot. A greater

response was obtained from a combination of the fermentation material and fish liver mycelium. The fish liver mycelium failed to stimulate growth when fed in the absence of either an antibiotic or the fermentation product. In a second trial the addition of orotic acid to a corn-soybean meal type ration produced a growth response comparable to that obtained from adding 3% of condensed fish solubles. A combination of the two supplements was no more effective in promoting growth than either alone. In a trial with corn-soybean meal-cottonseed meal rations, the addition of the above fermentation product elicited a growth response intermediate between the basal and that obtained from the addition of 3% fish solubles. A combination of fish solubles and fermentation product was only slightly more effective than the fish solubles alone.

PROTEIN AND FAT REQUIREMENTS OF BABY PIGS. E. R. Peo, Jr., G. C. Ashton, V. C. Speer and D. V. Catron, *Iowa State College.*

A 4 × 4 factorial experiment was conducted with 208 pigs to determine the protein and fat requirements of the baby pig. Protein levels of 15, 20, 25 and 30% were fed each in a reciprocal relationship with 0, 2.5, 5 and 10% fat. The protein used was composed of a 50–50 mixture of 50% solvent soybean oil meal and low-heat, spray dried skimmilk. Stabilized lard was used as a source of fat. Energy determinations were made on all rations with the bomb calorimeter. The pigs were removed from the sows at approximately one week of age (average weight 6.5 lb.) and were started immediately on dry experimental rations. The experiment was terminated after 49 days. Average total pig weight, average total gain and average feed per pound of gain are shown, respectively, for each level of protein and fat as follows: 35 day totals: *Protein*—(15%)—20.7, 13.6, 1.94; (20%)—23.3, 16.1, 1.66; (25%)—24.0, 15.2, 1.78; (30%)—23.0, 14.5, 1.67. *Fat*—(0%)—22.7, 15.8, 1.62; (2.5%)—22.8, 14.6, 1.81; (5.0%)—22.9, 15.2, 1.66; (10%)—22.5, 13.7, 1.82. Response to protein levels showed a quadratic statistical significance with the best overall performance shown by the pigs fed the 20% protein rations. There was no significant difference between the levels of fat fed, however, from a physical standpoint the advantages of adding fat to the rations outweighed any advantage obtained by excluding fat from the rations. These data also seem to indicate, as might be expected, that better results are obtained with the higher level of fat (10%) on the higher level of protein (30%). The 56 day data show similar trends.

THE EFFECT OF STILBESTROL AND TESTOSTERONE ON GROWTH AND CARCASS QUALITY OF SWINE. T. W. Perry, W. M. Beeson and F. N. Andrews, *Purdue University.*

Weanling Duroc pigs averaging 45 lb. liveweight were assigned to six dry lots of ten pigs each in an experiment designed to study the effects of adding hormones to the free choice protein supplement. The treatments were as follows: Lot I, *negative control,* free choice corn, minerals and protein supplement of soybean oil meal 50, meat scraps 40, dehydrated alfalfa meal 10, and B-vitamins; Lot II, *positive control,* same as Lot I plus 60 gm. Terramycin per ton protein supplement; Lot III, *negative control plus stilbestrol,* 2.5 mg. stilbestrol daily to 125 lb., then 5.0 mg. stilbestrol per pig daily; Lot IV, *positive control plus stilbestrol,* same levels of stilbestrol as in Lot III; Lot V, *negative control plus testosterone,* 20 mg. testosterone per pig daily to 125 lb., then 40 mg. testosterone; Lot VI, *positive control plus testosterone,* same levels of testosterone as in Lot V. After 91 days on treatment, the average daily gain, daily feed consumption and feed per 100 lb. gain figures were as follows: Lot I, 1.38, 5.41, 392.8; Lot II, 1.51, 5.44, 359.8; Lot III, 1.40, 5.00, 358.0; Lot IV, 1.49, 5.57, 374.4; Lot V, 1.44, 5.32, 369.0; Lot VI, 1.42,

5.30, 373.5. The addition of stilbestrol or testosterone exerted no significant improvement in growth rate or feed efficiency over that exerted by antibiotic feeding. The feeding of stilbestrol resulted in enlarged teats in both the males and females and resulted in swelling of the vulva in the case of females.

AMINO ACID SUPPLEMENTS FOR SWINE RATIONS. W. H. Pfander and L. F. Tribble, *University of Missouri.*

The level of lysine, methionine + cystine, and tryptophane needed in the rations of pigs from weaning to approximately 100 lb. has been studied with 196 Chester White pigs. Soybean oil meal, casein, liver meal and dried whole egg were used as the only sources of protein or in combination with each other and with corn. Other basal rations contained the above ingredients and/or tankage, fish meal, shorts and alfalfa leaf meal. Supplements of L-lysine HCl (0.1–0.25%) DL-methionine (0.04%) and DL-tryptophane (0.04%) were added to the basal ration alone and in combination. The lysine requirement (based on growth rate and feed efficiency) appears to vary with the protein content of the ration. Rations containing 12, 14, 16 and 18% crude protein were satisfactory if 4.25, 4.50, 5.0, and 5.5% of the protein was lysine. The methionine plus cystine and tryptophane requirements appear to be independent of the protein levels tested and were 3.5 and 1.0% of the protein, respectively. These studies indicate that some practical swine rations containing the levels of protein recommended by the National Research Council can be improved with small additions of lysine, methionine and tryptophane. However, the response obtained from a combination of amino acids was not usually as great as the response obtained from the addition of the most limiting amino acid.

THE EFFECTS OF A MANGANESE DEFICIENCY ON THE GROWTH AND DEVELOPMENT OF SWINE. M. P. Plumlee, D. M. Thrasher, W. M. Beeson, F. N. Andrews and H. E. Parker, *Purdue University.*

This is the fourth in a series of experiments to study the effect of low-manganese rations on growing fattening swine. In experiments 1 and 2 boar pigs and in experiment 3 female pigs were fed either purified or semi-purified rations containing from 1.0 to 40.0 p.p.m. Mn from weaning (35 lb.) to market weight. No differences between Mn levels were observed in either growth rate, feed efficiency, organ weights, blood phosphatase, or in Mn, Ca or P content of hock bones. The Mn content of several soft tissues was reduced from 3 to 5 fold in all animals on the low-Mn treatments. In the fourth experiment 20 crossbred female pigs were taken from the sow at three weeks of age (9.4 lb.), divided equally as to size and litter, and group fed a purified ration containing either 0.5 or 40.0 ppm, Mn to a weight of approximately 200 lb. Early weaned pigs were used to minimize the pre-experimental tissue Mn accumulation. No differences in weight gains or feed efficiency were noted between treatments. Backfat measurements, photographs, X-ray pictures, histological studies, tissue analyses, and visual observations showed the following conditions in the low Mn animals not present in the 40 ppm Mn group: significantly thicker backfat, shorter in body length and height, bowing of the front legs, enlargement of the knees and hocks, weakness in the hind legs and pasterns, partial closure of the epiphyseal plate, a marked narrowing of the epiphyseal cartilage of the distal end of the radius and ulna, and an extreme Mn depletion of soft tissues.

THE VALUE OF SEVERAL NON-PROTEIN NITROGEN COMPOUNDS AS PROTEIN SUBSTITUTES IN LAMB FATTENING RATIONS. W. W. Repp, W. H. Hale and Wise Burroughs, *Iowa State College.*

Research was initiated to study selected non-protein nitrogen (NPN) compounds as protein substitutes in ruminant rations realizing that the ideal compound should be non-toxic yet that its nitrogen should be readily available to rumen micro-organisms. Two group feeding and two individual feeding experiments were conducted using the following compounds: urea, ammonium acetate, ammonium formate, ammonium propionate, propionamide and formamide. In the group feeding trials and in one individual feeding trial all NPN compounds were added in amounts necessary to furnish 50% of the protein equivalent of the ration. Rates of gain with urea, ammonium acetate, ammonium formate and propionamide compared favorably with the conventional protein ration. As previous toxicity studies had shown that propionamide was non-toxic an individual feeding trial was conducted using this compound at two levels (15% and 30% of the protein equivalent) and control rations of urea at the same levels. Rates of gain on the urea and propionamide rations were about .50 lb. per lamb daily which was only slightly lower than gains on the conventional protein ration. In vitro studies to determine the rate at which sheep rumen micro-organisms adapted themselves to the release of propionamide nitrogen after receiving it in the ration indicate that full adaptation requires about three weeks.

ANTIBIOTICS AND METHOD OF FEEDING FOR BABY PIGS. W. M. Reynolds, G. P. Pendergrass, H. G. Luther, *Chas. Pfizer & Co., Inc.*

Studies have been conducted dealing with the effect of various rations and feeding methods on growth rate and feed efficiency of baby pigs raised off the sow. Investigations were also conducted evaluating (1) sources of carbohydrates, (2) substitution of dried skim milk for soybean oil meal, and (3) evaluation of purified diet consisting of an isolated soy-protein, cerelose and vitamin and mineral supplementation. The results from these trials established that the addition of Terramycin is of definite value in producing growth response, and that a dry feeding program reduces scouring, mortality and promotes more rapid gains than a milk feeding regime. The addition of a fermentation residue containing one or more of the yet unidentified growth factors to a purified diet improved the performance equal to or greater than diets containing high levels of dry skimmed milk.

PROTEIN REQUIREMENTS OF PREGNANT EWES. R. M. Richard, M. R. Light, D. W. Bolin, W. E. Dinusson and M. L. Buchanan, *North Dakota Agricultural College.*

Practical farm rations were used in studies designed to obtain information on the minimum protein requirements of pregnant ewes. One hundred pregnant ewes were divided equally on the basis of weight and age and wintered at different levels of protein intake. In comparison with ewes which received approximately one-half the recommended level of protein, ewes which were fed the recommended level (NRC, 1949) gained an average of 17.3 lb. more during pregnancy, dropped a higher percentage of live lambs (170 vs. 142 percent), and produced lambs which averaged 1.6 lb. heavier at birth. Post-partum lamb mortality was higher among lambs from ewes which received the lower level of protein during gestation. Also, reduction in protein fed during pregnancy resulted in decreased milk production as indicated by the rate of growth of lambs produced.

EFFECTS OF FEEDING HIGH LEVELS OF AMMONIATED MOLASSES TO DAIRY STEERS ON VARIOUS FEEDING REGIMES. L. L. Rusoff, F. T. Landagora, J. G. Lee, Jr. and J. E. Bertrand, *Louisiana Agricultural Experiment Station.*

Our previous work has demonstrated that a 10% level of ammoniated inverted molasses (32% protein equivalent) in a grain ration can be substituted satisfactorily for cottonseed meal on an equivalent N basis. Recently it has been reported by some investigators that ammoniated molasses causes a "stimulation" in animals. This study was made to determine whether levels above 10% in the grain ration, fed along with hay, would cause "stimulation." Young dairy steers weighing 180 lb. were "excited" at the 15% level while the 25% level caused stimulation in animals weighing 325 to 500 lb. It took 1.5 to 3.8 lb. of ammoniated molasses per 100 lb. of body weight to produce "excitability" which occurred within 2 to 5 days after the grain containing the molasses and lasted 1 to 3 hours. Non-protein N values of the blood prior to feeding the molasses and immediately following "excitability" were determined. No consistent variations in NPN values were found. The pH of the ammoniated molasses (pH 6, 7, or 8), and various roughage/grain ratios in iso-nitrogenous rations did not appear to be factors in causing the "stimulation." Ammonium carbonate administered by stomach tube or fed in blackstrap did not produce any "stimulation."

THE EFFECT OF AUREOMYCIN AND ARSANILIC ACID ON NITROGEN BALANCE IN PIGS. J. M. Russo, L. E. Hanson and J. J. Jezeski, *University of Minnesota.*

Seventeen balance trials have been completed to measure the effect of arsanilic acid on nitrogen retention in the growing pig. Seven similar trials were completed in which the effect of aureomycin on nitrogen retention was measured. The data to be reported will include that obtained in the last four trials. Four littermate Chester White barrows weighing approximately 50 lb. each at the beginning of the experiment were fed. Two served as controls and two were fed either arsanilic acid or aureomycin. The control diet was a practical mixture containing slightly more than 12% crude protein. Arsanilic acid was fed at a level of 60 grams, and aureomycin at a level of 20 grams per ton of feed. The experiment was designed so that during the four trials each pig of each pair was subjected to each treatment twice. A "t" test for each of the two pairs of animals revealed a highly significant difference in favor of increased nitrogen retention from the addition of either aureomycin or arsanilic acid to the control ration. In connection with these same four trials a study of samples of feces, stored at 0° F. immediately upon being voided, was made. Fecal samples for each pig and for each trial were separated into four fractions by filtration and differential centrifugation. Direct microscopic and plate counts of samples from each of these fractions indicated that marked concentration of bacteria into one of these fractions had been effected. Nitrogen determinations of each of the four fractions indicated that a reduction of nitrogen in this fraction appeared to be one of the primary causes of the increased nitrogen retention when aureomycin or arsanilic acid was added to the control ration.

AN EVALUATION OF CANE MOLASSES IN SWINE RATIONS WITH AND WITHOUT ANTIBIOTIC SUPPLEMENTATION. R. F. Sewell, W. C. Stringer and A. E. Cullison, *University of Georgia.*

The efficacy of cane molasses as a partial substitute for corn in swine rations was studied. Forty-two Duroc and Poland China pigs (average weight 77.8 lb.) were divided into six uniform groups and fed as follows: Lot 1—Basal; Lot 2—Basal

plus aureomycin; Lot 3—Medium level molasses; Lot 4—Same as 3 plus aureomycin; Lot 5—High level molasses and Lot 6—Same as 5 plus aureomycin. In lots 3 and 4 molasses replaced corn by weight at a 10% level of the ration initially. The molasses level of the rations was increased by 10% increments when the pigs averaged 100 lb. and 150 lb., respectively. Thus, levels of 10, 20 and 30% molasses were fed to these groups at the various stages of growth. A similar substitution was followed in Lots 5 and 6 except that a 20% level of molasses was fed initially and was increased to 30 and 40% levels as the pigs increased in weight. All rations were adjusted to an isonitrogenous basis. The average daily gain and feed conversion figures for the various lots were as follows: Lot 1—1.39, 446.0; Lot 2—1.64, 388.0; Lot 3—1.35, 538.0; Lot 4—1.61, 451.0; Lot 5—1.32, 503.0 and Lot 6—1.56, 462.0. Significantly (P equal to 0.05) faster growth rates were obtained on rations containing aureomycin. No significant differences were obtained between molasses levels. Under the conditions imposed, molasses appears to have a replacement value of 85 to 90% that of corn on a weight basis.

VITAMIN A REQUIREMENTS OF BABY PIGS. B. E. Sheffy, Nicholas Drouliscos, J. K. Loosli and J. P. Willman, *Cornell University*.

Two replicates of 12 one-day old littermate Yorkshire pigs were used in this study. A semi-synthetic vitamin A-low diet consisting of skimmilk, casein, lard, minerals and vitamins was fed. After a 4-week depletion period, supplementation levels of 0, 6, 12, and 18 mcg. of vitamin A acetate per kilogram body weight were administered orally daily. Growth, blood levels and liver storage were used as criteria of vitamin A sufficiency. Supplementation with 6 mcg./kg body weight will provide good growth and will prevent the appearance of gross deficiency symptoms. A level of 12 mcg./kg body weight can restore initial blood plasma levels of vitamin A. Supplementation with 18 mcg. can be considered to represent the minimum requirement to provide a trace of liver storage of vitamin A.

FURTHER OBSERVATIONS ON THE EFFECT OF SOURCE OF PROTEIN ON THE LAMB AND WOOL PRODUCTION OF MATURE RANGE EWES. S. B. Slen and F. Whiting, *Alberta Experimental Station*.

Six groups of mature range ewes (16 in each) were individually fed isocaloric rations containing six different sources of nitrogen,—linseed oil meal, soybean meal, meat meal, urea, alfalfa, and field peas to determine the effect of source of protein on lamb and wool production. The concentrate was fed in pelleted form and the roughage was fed chopped. Average feed consumption of the different lots was 3.2 lb. per head daily with the exception of lot 4 (urea) which was 2.8 lb. The rations, fed from 8 weeks before breeding until 6 weeks before lambing, contained an average of 7.7% protein on a dry matter basis (less than 50% of which was contributed by the ingredients in the basal ration). Those fed during the latter stages of pregnancy and during lactation contained an average of 10.8% protein (less than 40% of which was contributed by the ingredients in the basal ration). Significant differences (p less than .01) in wool production between lots were obtained. The ewes fed rations containing meat meal and urea produced significantly less wool than the other groups. However, the decrease in the urea group could be accounted for by a lowered feed consumption. Average birth weights of single and twin lambs from the ewes receiving the ration supplemented with urea were significantly lighter than those from the ewes receiving linseed oil meal and soybean meal. Average lamb weights at 8 weeks of age showed no significant differences between sources of protein with the exception of the urea group. These differences in lamb weights could be accounted for by the differences in feed consumption.

THE RELEASE OF AMMONIA NITROGEN FROM UREA, AMMONIATED MOLASSES AND SOYBEAN OIL MEAL IN THE PRESENCE OF RUMEN MICROORGANISMS. O. T. Stallcup, *University of Arkansas.*

Four series of experiments have been employed to determine the relative amounts of free ammonia nitrogen associated with urea, ammoniated molasses, 15.4% protein equivalent, ammoniated invert molasses, 33.3% protein equivalent and soybean oil meal in the presence of rumen microflora and the enzyme urease. In the first fermentation series urea and the two ammoniated molasses were compared by means of the artificial rumen technique. In the second fermentation series soybean oil meal and the two ammoniated invert molasses were compared by means of an artificial rumen. Urea, soybean oil meal, and the two ammoniated invert molasses were incubated for one hour at 40° C., with and without the presence of the enzyme urease in the media. In a fourth experiment, urea and the two ammoniated molasses were added to the rumen of a fistulated steer subsisting on a diet of Korean lespedeza hay. The level of free ammonia nitrogen in the rumen liquor was determined. Evidence is presented to indicate that there was some free ammonia in both samples of ammoniated molasses. The amount of free ammonia associated with these materials did not increase significantly with prolonged fermentation in an artificial rumen nor was there a significant amount released in the presence of the enzyme urease. Considerable amounts of ammonia nitrogen are released from urea in the presence of microflora both *in vitro* and in the presence of the enzyme urease. Small amounts of ammonia nitrogen are released from soybean oil meal in the presence of rumen microflora and the enzyme urease.

METHIONINE, LYSINE AND TRYPTOPHAN SUPPLEMENTATION OF GROWING FATTENING RATIONS FOR SWINE. E. L. Stephenson, P. R. Noland, J. J. Miner and W. B. Clower, *University of Arkansas.*

Growing fattening pigs were fed rations containing cottonseed meal and a combination of cottonseed meal and soybean oil meal as the respective protein supplements. Lysine was found to be a limiting amino acid in all rations whereas both lysine and tryptophan were deficient in the feed containing cottonseed meal as the principle protein supplement. In supplementing the ration the most effective level of dl lysine monohydrochloride was 0.1%. Higher levels of this amino acid appeared to depress growth and lower levels failed to promote a maximum response. The optimum supplementary level of dl tryptophan was found to be 0.02%. The cottonseed meal used in this study was prepared by the pre-press solvent process and had a free gossypol content of .036%.

EFFECTS OF GESTATION AND LACTATION DIETS ON LIVABILITY OF PIGS. J. W. Stevenson, N. R. Ellis and Robert J. Davey, *U. S. Department of Agriculture.*

In the first phase of the experiment (1949–50) a basal diet made up of yellow corn, oats, tankage, soybean meal, linseed meal, alfalfa meal, and minerals was not improved, as measured by reproductive performance, by supplementation with riboflavin, pantothenic acid, and condensed fish solubles. Results of the second series (1951) indicated an increased viability of suckling pigs due to the addition of alfalfa meal (15%) to a corn-soybean-mineral gestation diet and an increase in average weaned weight of pigs attributed to presence of aureomycin-vitamin B_{12} concentrate in the lactation diet. Significance was not established due to the limited size of the test although differences were substantial. In the third phase of the experiment (1952–53) the addition of an antibiotic-B_{12} supplement to a basal gestation diet containing 15% crude protein of both plant and animal origin significantly

improved pig survival rate (P less than .01). Nearly identical improvement (approximately 10%) resulted from raising the crude protein content of the gestation diet to 20%. The addition of the antibiotic-B_{12} supplement to the 20% gestation diet did not significantly improve pig survival. Suckling pigs which received a pelleted diet (containing antibiotic-B_{12} supplement) provided in a creep were significantly (approx. 8 lb.) heavier at 56 days than pigs not receiving this supplemental feed (P less than .01).

THE EFFECT OF GROUND SUN-CURED ALFALFA ON THE DRY LOT REPRODUCTIVE PERFORMANCE OF SWINE. H. S. Teague and W. L. Robison, *Ohio Agricultural Experiment Station.*

Eighty-four gilts and 8 sows were employed in four dry lot experiments to determine the effect of sun-cured alfalfa on reproductive performance. Either an 18% alfalfa diet or one containing no alfalfa or other legume was fed. Dietary crude fiber was equalized by employing ground ear corn in the no-alfalfa diet and sufficient vitamins A, D, E, and 8 B-vitamins were added to more than supply the calculated amounts of these vitamins furnished by 18% alfalfa. The diets were fed for at least 19 days prior to breeding and during gestation. Eight sows in the second, 10 gilts in the third, and 16 gilts in the fourth experiment were sacrificed early in gestation to observe the possible effect of diet on ovulation rate and embryonic survival. All other females were allowed to farrow. Alfalfa-fed gilts farrowed an average of 9.83 live and 0.87 stillborn pigs compared to 8.64 live and 0.96 stillborn farrowed by those not fed alfalfa. Neither vigor at birth, as classified, nor average birth weight was affected by the maternal diets. The mean number of corpora lutea in sows fed the alfalfa diet was 13.0 and in gilts, fed the same diet, 13.5. Sows fed the no-alfalfa diet possessed an average of 12.0 and gilts 11.9. The results suggest that a factor or factors in alfalfa primarily affected ovulation rate and not prenatal growth or survival.

THE EFFECT OF ALFALFA ASH AND CERTAIN OF ITS MINERAL CONSTITUENTS ON THE UTILIZATION OF COTTONSEED HULLS BY SHEEP. C. W. Tefft, A. D. Tillman, R. J. Sirny and Robert MacVicar, *Oklahoma Agricultural Experiment Station.*

Previous work at this laboratory showed that alfalfa ash improved the utilization by sheep of a semi-purified diet containing 60% cottonseed hulls. Later it was found that equally beneficial effects were obtained by feeding a "synthetic" alfalfa ash compounded from inorganic salts to resemble the mineral composition of natural alfalfa ash. Two experiments involving 61 lambs in a "depletion-repletion" regimen were designed to determine the mineral(s) contained in alfalfa ash responsible for the increased utilization of cottonseed hulls. During the depletion phase, all animals were fed the semi-purified basal diet for at least 28 days and a marked loss in weight was observed. During the 21-day repletion phase which followed immediately, the effect of omitting certain minerals from the synthetic alfalfa ash upon the growth of the depleted sheep was studied. When cobalt, magnesium, manganese, or zinc were individually omitted, no depression in growth was noted. When iron or copper was omitted a depression in growth occurred with a greater depression being noted in the case of iron. Omission of molybdenum gave divergent results in the two trials. In a growth trial with 20 lambs, the addition of alfalfa ash to a ration containing equal quantities of cottonseed hulls and a concentrate mixture composed of natural feedstuffs resulted in an increased gain of 23% with feed efficiency being increased by 32%.

EFFECT OF AUREOMYCIN ON DIGESTION OF LOW QUALITY ROUGHAGE BY SHEEP. C. M. Thompson and R. B. Grainger, *University of Kentucky.*

A series of three metabolism trials consisting of three groups of two wethers each was conducted with 18 wether lambs fed a daily ration composed of 640 gm. ground corn cobs; 116.88 soybean oil meal; 16 alfalfa meal; 16 molasses; minerals (Ca P NaCl Co) and cod liver oil. The wethers in Group I received the basal ration: Group II as I plus 10 mg. pure crystalline aureomycin-HCl per pound of ration, and Group III as I plus Aurofac 2A to furnish 10 mg. aureomycin-HCl per pound of ration. The wethers were placed in metabolism cages and after becoming adjusted to surroundings and rations, a 10-day preliminary period was followed immediately by a 10-day collection period. The wethers in Groups II and III exhibited anorexia within 48 to 72 hours after antibiotic supplementation. Appetites returned to normal within two or three days. The average apparent digestion coefficients for crude fiber was 71.4, 60.4, and 59.7% for the wethers in Groups I, II, and III respectively. These differences are statistically significant. There was no difference in nitrogen retention among groups. Although the apparent digestibility of dry matter, nitrogen-free extract, and energy was lower for the wethers on supplemented rations the differences were not statistically significant.

THE EFFECT OF VARIOUS CARBON SUBSTANCES ON *IN VITRO* ACTIVITY OF SHEEP RUMEN MICROORGANISMS. W. J. Tyznik, O. G. Bentley, H. H. Weiser, T. S. Sutton, A. L. Moxon and Assistants, *Ohio Agricultural Experiment Station.*

Sheep rumen studies, using the artificial rumen technique, were conducted to determine the effect of various carbon compounds on cellulose digestion, nitrogen fixation and volatile fatty acid production. The amount of carbon from various compounds was based on the equivalent of 4.5 gm. of cellulose (Solka flok). All carbon compounds were used at levels of 25%, 50%, 75% and 100% of the carbon present in cellulose and activity compared to cellulose alone. Starch and maltose increased cellulose digestion, nitrogen fixation and total volatile fatty acid production with a 100% increase in the proportion of butyric acid produced. Glucose when used alone or in combination with cellulose gave the same relative proportion of volatile fatty acids as cellulose alone but significantly increased the total acid production. Glucose was not as stimulatory to cellulose digestion or nitrogen fixation as starch or maltose. Lactic acid increased the proportion of propionic acid and total volatile acids but reduced nitrogen fixation and cellulose digestion when used at a concentration greater than 25% of the total carbon. Sodium pyruvate had no effect on cellulose digestion or nitrogen fixation when it replaced more than 25% of the carbon but was stimulatory below the 25% level. The addition of starch, maltose, lactic acid and sodium pyruvate yielded a varying amount of valeric acid (1%–7.5%). Addition of acetic, propionic and butyric acid to replace 25% of the carbon had no effect on cellulose digestion or total volatile fatty acid production. Acetic and propionic acid decreased cellulose digestion and nitrogen fixation whereas butyric acid had no effect.

LEVELS OF PANTOTHENIC ACID AND REPRODUCTIVE PERFORMANCE OF SOWS. D. E. Ullrey, D. E. Becker, S. W. Terrill and R. A. Notzold, *University of Illinois.*

Forty gilts were fed a semi-synthetic, pantothenic acid deficient ration for one month. Eight gilts were removed from test and the remaining thirty-two allotted to two replicates with four levels of pantothenic acid fed in each replicate. The total pantothenic acid contents of the diets fed were 0.5, 2.5, 5.5 and 8.5 mg. per

pound of feed. After a month of treatment, the gilts were bred. Reproductive performance of the gilts receiving the 0.5 and 2.5 mg. levels of the vitamin was unsatisfactory. No pigs were born on the lowest level, and some of the pigs born on the second level exhibited irreversible deficiency symptoms. Diarrhea, locomotor incoordination, and a persistent tremor were frequently noted. Several pigs were observed "goose-stepping" at three days of age. The reproductive systems of gilts receiving 0.5 mg. of pantothenic acid commonly appeared infantile. Ovaries were atrophic and inactive. Those individuals most severely affected by the deficiency exhibited increased blood pyruvate in the terminal stages. Milk samples obtained between the third and fifth week of lactation were assayed for free and bound pantothenic acid.

THE EFFECT OF ARSANILIC ACID AND 3-NITRO–4-HYDROXYPHENYLARSONIC ACID ON SELENIUM TOXICITY IN PIGS. Richard C. Wahlstrom, L. D. Kamstra and Oscar E. Olson, *South Dakota State College.*

Two experiments have been conducted to study the effect of arsanilic acid and 3-nitro–4-hydroxyphenylarsonic acid on selenium poisoning in the pig. A corn-soybean meal-tankage ration fortified with vitamins and minerals was used as the basal ration in both trials. In the first trial thirty weanling pigs were allocated to six treatments (1—Basal; 2—Basal + .01% arsanilic acid; 3—Basal + .005% 3-nitro–4-hydroxyphenylarsonic acid; 4—As 1 + 7 ppm selenium (as sodium selenite); 5—As 2 + 7 ppm selenium; 6—As 3 + 7 ppm selenium). The mean daily gains of the pigs on the treatments for an average of 105 days were 1—1.53; 2—1.59; 3—1.64; 4—1.22; 5—1.36; 6—1.64. One pig died in Lot 4 due to selenium poisoning and two of the remaining pigs showed definite selenium toxicity symptoms. No visible selenium toxicity symptoms were apparent in Lots 5 and 6. In the second trial five lots of 8 pigs each were fed the same basal ration as in trial one plus 10 ppm of selenium (as sodium selenite). The treatments and their respective daily gains were: (1) Seleniferous Basal, 0.86; (2) as 1 + .02% arsanilic acid, 1.43; (3) as 1 + .01% arsanilic acid, 1.25; (4) as 1 + .005% 3-nitro–4-hydroxyphenylarsonic acid, 1.33; (5) as 1 + .0025% 3-nitro–4-hydroxyphenylarsonic acid, 0.96. The higher levels of each of these organic arsenicals appeared to effectively counteract the toxic effects of selenium as noted in Lot 1.

COBALT POLYCYTHEMIA IN CATTLE. George M. Ward, C. F. Huffman and C. W. Duncan, *Michigan State College.*

Fourteen dairy calves ranging from 12 to 75 days of age were alloted to four groups. One group received no supplement but the remaining groups received daily supplements of either (a) cobalt sulfate at the rate of 5 mg. per pound of body weight, (b) sulfaguanidine at the rate of 2% of the dry matter intake, or (c) a combination of the two supplements. The red blood cell count reached 20 million per cubic millimeter in 3 of 4 calves on the cobalt supplement alone. Concurrent increases also occurred at a slower rate in hemoglobin concentrations and red blood cell volumes, which finally reached equivalent levels ranging from 15 to 20 grams percent and 46 to 57%, respectively. Sulfaguanidine supplementation produced no apparent change in the blood picture. Cobalt supplementation following or in conjunction with sulfaguanidine administration resulted in concurrent and equivalent increases in red blood cell counts, hemoglobin concentrations, and red blood cell volumes, but the increases were not as great as those encountered in the calves supplemented with cobalt alone. The severity of the polycythemia decreased as the animals approached 150 days of age. Subnormal weight gains were encountered in all calves receiving the cobalt supplement whereas the control calves gained at a

rate in excess of Ragsdale's standard. No response to cobalt supplementation was noted in a group of yearling heifers nor was any difference found in the rate of gain in body weight between the cobalt supplemented and control heifers. It was concluded that cobalt is able to produce polycythemia in calves before the rumen microflora is completely established.

DRIED RUMEN CONTENTS IN CALF MILK REPLACEMENTS. J. B. Williams and C. Jensen, *North Dakota Agricultural Experiment Station.*

The effect of adding artificially dried rumen contents to calf milk replacement rations was studied with 24 four day old dairy calves from the Holstein and Guernsey breeds, both male and female, and divided into three experimental groups with one whole milk control group. Group I was fed a replacer containing 2% dried rumen contents, 2% soybean lecithin and 2% aureomycin-B_{12} supplement; Group II ration contained the same ingredients as Group I except for aureomycin-B_{12} supplement; Group III ration contained lecithin and aureomycin-B_{12} supplement; and the control group got 400 lb. of whole milk. Two additional calves were fed a dried skim milk replacer containing 20% finely ground beet pulp. The average daily gains from birth to the end of ten weeks were as follows: Group I—0.71 lb.; Group II—0.69 lb.; Group III—0.65 lb.; control—1.43 lb. The dried beet pulp calves gained 1.06 lb. per day but showed some hair loss over the neck and shoulders at 4 weeks of age. The soybean lecithin was added as a dispersion agent and a source of fat but it did not prevent the milk replacers from settling out. The dried rumen contents with or without aureomycin-B_{12} supplement did not increase growth and the antibiotics alone did not increase growth.

Pasture and Forages Section

W. C. Weir, *Chairman*

THE APPLICABILITY OF THE FECAL CHROMOGEN TECHNIQUE IN DIGESTION STUDIES WITH SOME SOUTHERN-GROWN FORAGES. W. B. Anthony, C. M. Martin, J. G. Starling, L. Brown and E. L. Mayton, *Alabama Polytechnic Institute.*

Digestion trials were conducted with 4 forage crops commonly grown in the South in order to test the applicability of the fecal chromogen technique for digestibility as defined by Reid, et al. Coastal Bermuda, Pensacola Bahia grass, fescue and Lespedeza Serecia were studied. The experimental procedure consisted of feeding fresh-cut forage twice daily to yearling Hereford steers maintained in individual pens fitted with facilities for quantitative feeding. Steers received test forage for a preliminary period of 6 days and a test period of 4 days. For the 4-day test period animals were fitted with harnesses and collection bags, and fecal excretions were quantitatively collected. Digestion coefficients estimated by the fecal chromogen technique developed by Reid closely approximated coefficients obtained by the conventional method. Absorption spectral data for forage and feces extracts were similar for all 3 forages and reveal common isobestic points near 406 mu. For these forages the fecal chromogen technique (Reid, et al.) is a reliable method for estimating forage digestibility. Furthermore, A.M. and P.M. collections were alike with respect to relative chromogen concentration, and "grab" samples collected either in the morning or afternoon were satisfactory for determining dry matter digestibility by the chromogen technique.

RATION DIGESTIBILITY, GRAZING BEHAVIOR AND PERFORMANCE OF BEEF CATTLE AS AFFECTED BY SUPPLEMENTAL FEEDING ON WINTER PASTURE. D. M. Baird and O. E. Sell, *Georgia Experiment Station.*

Improving beef cattle performance by supplemental feeding on winter temporary pasture has not been generally successful when compared with cattle on the pasture alone. A test with yearlings included the three following treatments: (1) Pasture alone, (2) One-half feed grain ration on pasture and (3) Full fed grain ration on pasture. Some of the criteria used to evaluate the feeding methods included forage and total ration intake and digestibility, rate of gain and grazing behavior studies. Grazing behavior observations were made during continuous 24 hour periods. A.D.G. for the above three lots were 2.17, 2.32 and 2.35 lb. respectively. Differences in forage consumption were small between lots, with the full fed cattle consuming approximately 2 lb. less forage dry matter daily than cattle receiving pasture alone. Most striking and consistent was the lowering of digestibility of the total ration as increasing amounts of grain were fed on pasture. The average total ration digestibility was 71% for pasture alone and 67% for the full fed grain group on pasture. Digestibility and forage intake decreased with pasture maturity. The digestibility of high quality pasture (as high as 31% CP) was apparently depressed by the grain ration of corn and oats. The average 24 hour grazing time was 564, 450 and 377 minutes daily for the pasture, ½, and the full fed lots respectively. The $\frac{\text{ruminating time}}{\text{Grazing time}}$ (ratio) was .58, .84, and 1.03 respectively for the above lots. As the amount of grain was increased, the ruminating time increased. To properly supplement cattle on this type pasture the supplement must be both high in quality and energy with less emphasis on protein needs.

PERFORMANCE AND PARASITISM OF BEEF CALVES AS RELATED TO LEVEL OF PROTEIN AND PASTURE FEEDING. D. M. Baird, H. H. Vegors, O. E. Sell and T. B. Stewart, *Georgia Experiment Station—U. S. Department of Agriculture.*

The relation of animal performance and internal parasitism to ration protein level and amount of pasture forage available was studied. Beef calves were placed in two paddocks each of: (1) Drylot, (2) Semi-drylot (limited grass for parasite pickup), and (3) Abundant winter temporary pasture. Two levels of protein, approximately 20% above and below N.R.C. standards, but equated for T.D.N., were fed to calves in each of the above treatments. Calves comparable in age, weight, and fecal egg counts, were posted at the end of tests for parasite recovery. Forage intake of the pasture group was determined by the chromic oxide method. The A.D.G. for the above lots were 1.70, 1.91, and 2.31 ** lb. respectively; and 2.00 and 1.95 lb. daily for the grouped high and low protein lots. An inverse relation was found between worms and larvae. In the above three groups 10,768, 8,544 and 5,739 adult worms were recovered while 25, 369 ** and 1,926 ** larvae respectively were recovered per head. For the grouped high and low protein lots the adult worm recoveries were 7,671 and 9,030 * and recoveries for larvae were 300 and 1,247 * respectively. The level of protein fed had no effect on the forage consumption by the pasture cattle, thus it was apparently protein rather than forage consumption level that affected parasite load. When high vs. low protein groups were compared it was found that the low protein group of calves had significantly higher loads of larvae and adult worms, but these levels were not sufficiently high to impair growth rate. High protein in the drylot rations was beneficial but of no value on pasture with respect to cattle performance. (* Equals probability of 0.05; ** equals probability of 0.01.)

BLOAT STUDIES AND OBSERVATIONS. B. F. Barrentine, C. B. Shawver and L. W. Williams, *Mississippi Experiment Station.*

Paddocks of pure ladino clover were used in this study. Steers grazed the clover for a two hour period, morning and afternoon. At the end of the two hour period, the steers were checked for bloat and put in dry-lot with shade and water. Later in the study, a 1½ hour grazing period was used morning and afternoon which was equally satisfactory. Forty grade steers were used from which 14 good bloaters were selected. A total of 1575 cases of bloat were observed, seven of which were acute. A trocar or knife appears to be the only treatment of value in an acute case. The other cases of bloat recovered without treatment, even though many of the steers were extremely distended. Tympanometer readings on bloated steers indicated pressures of 20 to 60 mm Hg. In acutely bloated steers, the pressure was around 80 mm Hg. Death is apparently caused by tissue anoxia and asphyxiation. Death usually occurred after the steer had been bloated one to two hours. Twenty grams of pure methyl silicone given in capsules before grazing in the morning reduced the incidence of bloat for that period, but had no effect that afternoon. Treatment of bloated animals with preparations of methyl silicone appeared to be of doubtful value. Six grams of aerosol appeared to be without value in the prevention of bloat. The moisture content or lushness of growth of the clover did not appear to be a factor in the incidence or severity of bloat.

MEASUREMENT OF NUTRIENT LOSSES FROM GRASS SILAGE. J. W. Bratzler, R. L. Cowan, E. Keck and R. W. Swift, *Pennsylvania State University.*

First-cutting, unwilted, legume-grass forage (largely clover) was ensiled in two 5′ × 10′ wood stave silos. The treatments were: (1) with the addition of 112 lb. of molasses per ton of forage and (2) without the addition of any preservative. A sufficient quantity of the same unwilted forage was frozen immediately in order to provide forage as near the fresh state as possible for the subsequent determination of: (1) the feeding value of the forage itself and (2) the feeding value of the forage plus molasses at the same rate that it was added to the ensiled forage. Four digestion experiments with the same four sheep in each formed the basis for the determination of the nutritive value of the two silages, the forage itself, and the forage plus molasses. The T. D. N. values for the frozen forage, frozen forage plus molasses, no-preservative silage, and molasses silage were: 63.8, 66.6, 60.5 and 64.9 lb./100 lb. D. M., respectively. Losses of dry matter and energy from the green forage ensiled with molasses and without the addition of any preservative were 20.2 and 20.7%, and 21.8 and 22.4%, respectively, whereas the losses of digestible energy and T. D. N. were 24.2 and 22.2% and 29.5 and 25.8%, respectively.

COMPARISON OF ROUGHAGES FOR WINTER FEEDING AND MAINTENANCE OF BEEF CATTLE. Paul B. Brown, R. A. Damon, Jr., C. B. Singletary and E. H. Vernon, *Louisiana State University.*

The purpose of this experiment was to compare the feeding value of sugar cane bagasse, native grass hay, rice straw and cottonseed hulls when included in the wintering rations of beef cattle. Thirty-two head of good grade beef steers averaging 656 lb. in weight were equally divided into four lots on the basis of breed, condition, and weight and fed the experimental ration free choice for a period of 154 days. In addition to the roughages, which formed 62.5% of the ration, the following concentrate feeds were added on a percentage basis; blackstrap molasses 20%, cottonseed meal (41% protein) 11%, alfalfa meal 5%, bone meal 1%, and salt 5%. Each roughage was ground on a hammer mill, mixed with

the other dry ingredients and then blended with the molasses under pressure. The average daily gain, average daily feed consumption and feed required per pound of gain for the respective rations were as follows: sugar cane bagasse—.88 lb., 17.29 lb., and 19.65 lb.; native grass hay—1.51 lb., 26.71 lb., and 17.69 lb.; rice straw—1.88 lb., 18.29 lb., and 15.05 lb.; and cottonseed hulls—1.49 lb., 30.52 lb., and 20.48 lb. The differences in daily gain between Lot 1 and the other three lots were significant. The differences between Lots 2, 3, and 4 were not significant.

THE COMPARATIVE FEEDING VALUE OF BROMEGRASS HAY AND SILAGE DETERMINED BY FEEDING TRIALS AND DIGESTION STUDIES. A. L. Brundage and W. J. Sweetman, *Alaska Agricultural Experiment Station.*

Digestion trials, employing the chromium oxide technique, have been conducted over the past two years to compare the relative effect of barn drying and ensiling on the nutritive value of identical cuttings of smooth bromegrass. The T.D.N. contents were 64.93% and 56.27% for the hay and silage respectively. The average percent composition and digestion coefficients of the hay and silage on a dry matter basis were: protein—13.0 and 62.91, 13.1 and 45.94; ether extract—2.4 and 47.10, 3.2 and 62.16; crude fiber—29.6 and 67.97, 33.0 and 64.34; N.F.E.—46.6 and 73.16, 40.2 and 61.04. Calculated T. D. N. values similar to those above were obtained in 1953–54 when lactating dairy cows were fed roughage at a constant dry matter intake of 2 lb. per 100 lb. body weight. In the four previous years, however, when roughage was fed ad libitum, silage had a higher calculated T.D.N. than hay, largely the result of lower silage consumption. The daily milk production over all 30 day experimental periods was averaged and the linear regression formula calculated for each roughage. The regression coefficients (rates of decline) were greater for the silage group than for the hay group when all 30 day lactations, initial lactations, or lactations following the opposite roughage were considered. These differences were all highly significant (P less than .01). It is proposed that consideration of rate of decline will be of value in resolving the discrepancy between the feeding trial and digestion trial data.

A COMPARISON OF ELEPHANT, PARA, PANGOLA AND GUINEA GRASS PASTURES FOR NATIVE AND NATIVE × BRAHMAN YEARLING BULLS. Irwin A. Dyer, Floyd R. Olive and Ernesto Nunez, *Centro Nacional de Agronomia, El Salvador.*

Thirty two yearling bulls were alloted to a randomized block experiment of 16 treatments for 24 weeks in which the comparative value of elephant, para, pangola and guinea grasses was studied. Treatments were: 4 lots, no treatment; 4 lots, nitrogen fertilization; 4 lots, trace mineral supplementation ($CuSo_4$, $MnSo_4$ and $ZnSo_4$); and 4 lots with nitrogen fertilization and trace mineral supplementation. One Native and one Native × Brahman crossbred bull were in each of the ½ manzana (0.86 acre) lots. Animals were rotated each six weeks so that at the end of the 24 weeks each animal had grazed each of the 4 grasses but had remained on the same treatment with regard to nitrogen fertilization and trace mineral supplementation. All animals had free access to a 33.3% bonemeal, 66.6% salt mixture. The average daily gain, in pounds, for the Native bulls was 0.57 while that of the Native × Brahman was 0.69. The gains produced by the different grasses and treatments were: elephant grass—0.14; guinea—0.12; para 1.24; pangola 1.54; no treatment 0.36; nitrogen fertilization 0.55; trace minerals supplementation 0.76; and trace mineral supplementation and nitrogen fertiliza-

tion 0.88. Differences in grasses were highly significant as were the differences in treatments. This test was conducted during the dry season, consequently, the gains are not representative of the entire year.

YIELDS OF HOLOCELLULOSE PREPARED FROM FECES BY ACID CHLORITE TREATMENT. R. E. Ely, H. G. Wiseman, H. M. Irvin and L. A. Moore, *U. S. Department of Agriculture.*

Holocellulose was prepared from feces samples of 10 cows fed different forages. The same acid chlorite treatment was used that gave satisfactory results for the preparation of holocellulose from the various forages fed these animals. Yields of holocellulose from six of the feces samples were approximately equal to the theoretical content. Four of the feces samples gave holocellulose yields (ash, protein and lignin-free) greater than the theoretical yield. Comparison of the reducing sugar contents of the extractive-free feces and the respective holocellulose samples indicate that a non-carbohydrate constituent increases the holocellulose yield of these samples. Holocellulose samples prepared from feces had higher apparent lignin contents than the holocellulose samples prepared from the respective forages.

A PRELIMINARY REPORT ON A ZERO PASTURE PROGRAM FOR DAIRY CATTLE. B. W. Henderson, Jr. and J. W. Cobble, *Rhode Island Agricultural Experiment Station.*

Dairymen have given attention in recent years to a system of pasture utilization called zero pasture in which the pasture crop is field chopped and fed to cows in the barn or on dry lot. In an attempt to determine the advisibility of such a pasture utilization program, two groups of 14 cows each were selected, as nearly alike as possible in breed, body weight, stage of lactation and production of fat corrected milk. One group was handled in the conventional manner on pasture, the other group was dry lot fed the pasture crop which was field chopped. Daily records were kept for the latter group of average daily consumption of green forage and dry matter content of forage fed; and for both groups changes in body weight and production of fat corrected milk. Average daily consumption of green feed per head varied from 106 to 208 lb., dry matter varied from 9.4 to 28.0% No significant changes were noted in body weight. Daily milk production was more consistent for the zero pasture group.

GRASS SILAGES AS THE ONLY SOURCE OF NUTRIENTS FOR MILKING COWS. D. L. Hill, B. J. Walker and N. S. Lundquist, *Purdue University.*

Eighteen milking cows were divided into two groups of nine cows each and were fed winter roughages as the sole source of nutrients for 147 days. Group I was fed first-cutting legume-grass silage and Group II was fed first-cutting hay harvested from the same field at the same time. Both forages were harvested when the alfalfa was in the bud stage. The cows were fed according to individual appetite and had free access to a mineral mixture consisting of equal parts of bonemeal and mineralized salt. The average daily dry matter consumption per cow in group I was 30.5 lb. compared to 29.7 lb. for group II. The average daily milk production (F.C.M.) per cow was 23.1 lb. for group I compared to 20.4 lb. for group II. The average change in body weight per cow during the experiment was + 63 lb. for group I and + 26 lb. for group II. Considerable variation in individual cow performance was noted within groups. The individual average daily silage consumption (Group I) ranged from 78.6 lb. to 144.8 lb. while the range in hay consumption (Group II) was 19.1 lb. to 44.4 lb. The corresponding

ranges in milk production (F.C.M.) for groups I and II were 16.2 lb. to 28.9 lb. and 14.1 lb. to 30.6 lb., respectively. The data obtained support the thesis that high quality winter roughages can be used to a greater extent in feeding milking cows.

LAMB RESPONSES TO ANTIBIOTICS AND PELLETED RATIONS. R. M. Jordan, *South Dakota State College.*

The effect of feeding aureomycin and pelleting the ration of fattening lambs was studied using 4 lots of 12 lambs each. The lambs were shorn prior to being placed on experiment and had access to fresh water, mineral and shelter. They were full fed as follows: Lot 1: Long alfalfa hay and shelled corn, equal parts; Lot 2: Ground alfalfa hay and corn in equal proportion mixed and pelleted plus .3 lb. of long alfalfa per lamb daily; Lot 3: Same ration as fed Lot 2, plus 10 mg. of aureomycin per pound of pelleted ration; Lot 4: Three parts of ground alfalfa hay and one part ground corn, plus 10 mg. of aureomycin per pound of mix. This mixture was pelleted and fed with .3 lb. of long alfalfa per lamb daily. Average daily rates of gain were, 0.51, 0.54, 0.60 and 0.57 lb. for lots 1, 2, 3 and 4 respectively. Pelleting the ration or adding aureomycin did not increase feed consumption. Total feed consumption per lamb was the greatest in Lot 4, but corn consumption was the lowest. Pelleting the ration failed to increase feed efficiency and increased feed costs materially. Lambs in Lot 3 (aureomycin) required 10% less feed per 100 lb. of gain than Lot 2.

FECAL CHROMIC OXIDE CONCENTRATION IN 12 DAIRY COWS AS RELATED TO TIME AND FREQUENCY OF ADMINISTRATION AND TO FEEDING SCHEDULE. W. N. Linkous, W. A. Hardison, G. C. Graf and R. W. Engel, *Virginia Agricultural Experiment Station.*

Fecal samples were obtained at 2 hour intervals for 120 consecutive hours from each of 12 lactating animals. The animals were divided into 4 treatment groups of 3 animals each and were fed twice daily on grain, hay and silage. The animals each received 15 gm. of Cr_2O_3 daily in capsule form. Group I received 15 gm. of Cr_2O_3 at the morning feeding; group II received 7.5 gm. of the indicator at the morning feeding and 7.5 gm. at the evening feeding; group III received 15 gm. midway between the morning and evening feedings; and group IV received 7.5 gm. midway between the morning and evening feedings and the other 7.5 gm. between the evening and morning feedings. Considerable variation was observed in the Cr_2O_3 concentration in the feces. Greatest concentration occurred at the intervals 12 noon to 2 p.m. and 2 a.m. to 4 a.m. The time interval between these points corresponded with the time interval between evening and morning feeding, indicating a relationship between feeding time and indicator concentration in the feces. The most reliable estimate of total fecal D.M. output was obtained by compounding samples taken at 6–8 a.m. and 6–8 p.m. This is in general agreement with other published results. Individual cow variations were large suggesting that more animals and fewer collection days would yield more reliable information with this indicator than limited animal numbers and more collection days.

HAY CROP SILAGE VS. SILAGE AND HAY FOR MILK PRODUCTION. R. H. Lush, A. G. Van Horn and W. M. Whitaker, *University of Tennessee and U. S. Department of Agriculture.*

Three reversal silage vs. silage and hay feeding tests with groups of Jersey cows have been conducted at the Dairy Experiment Station, Lewisburg. Good

quality alfalfa-orchardgrass silage was compared to varying amounts of silage and mixed legume hay on a dry matter basis. Grain was fed according to milk production. In the first test, the cows on all silage consumed an average of 57.8 lb. silage daily and produced 4.3% more 4% F.C.M. than for periods on 8 lb. No. 3 lespedeza hay and 34.4 lb. silage. However, 7.4% more nutrients were consumed during the all-silage periods. In the second test, the cows ate an average of 54.7 lb. silage, the same rate of T.D.N.'s but produced 2% less milk than for the 8.9 lb. of alfalfa-orchardgrass hay and 31.9 lb. silage consumed daily. In the third experiment, cows on all-silage ration consumed 56.4 lb. silage and produced 22.4 lb. milk daily; for a new rate of three-quarters silage periods an average of 45.5 lb. silage and 4.6 lb. hay was consumed and 24.3 lb. milk produced daily; and for the other periods, an average of 31.5 lb. of silage and 9.3 lb. of hay, giving an average of 23.0 lb. 4% F.C.M. daily. The hay periods resulted in over 2% higher T.D.N. intake and slightly more milk than for the all-silage periods. The three winters results show 3.8% less T.D.N. consumption for one-half silage + one-half hay but .22% more 4% F.C.M. than for all silage periods. The last year indicates 5.7% more milk where three-quarters silage-on-one-quarter hay was fed over all-silage. There was little difference in liveweight changes.

FIELD USE OF THE CHROMIC OXIDE AND FECAL CHROMOGEN TECHNIQUES IN EVALUATING PASTURE HERBAGE WITH GRAZING ANIMALS. C. M. Martin, W. B. Anthony and J. G. Starling, *Alabama Polytechnic Institute.*

The digestibility and intake of common and coastal Bermuda and Pensacola Bahia grass have been measured with grazing steers by the chromogen-chromic oxide techniques. The experimental design was randomized complete block with two replications. Common Bermuda and Pensacola Bahia grass received 0, 80, and 160 lb. nitrogen per acre applied in three split applications during the growing season. Coastal Bermuda grass had an additional treatment of 320 lb. of nitrogen per acre. Increments of N fertilizer significantly increased the yields of digestible dry matter by all three grasses. Coastal Bermuda out-yielded the other grasses while Pensacola Bahia grass was intermediate. No statistically significant differences were found among the three forages for daily dry matter intake, N level and dry matter intake, or gain per unit of dry matter consumed. Nitrogen fertilizer improved forage digestibility. 7.79 lb. of forage dry matter produced a pound of gain in the first part of the grazing season; 20 lb. were required in late season. Both dry matter intake and digestibility were lowered as the grazing season progressed. The most important difference existing among the three forages for the production of beef is yield of forage per se and the response to N fertilizer Coastal Bermuda showed the greatest response to N.

THE VALUE OF GRASS SILAGE IN THE DIET OF THE YOUNG DAIRY CALF. G. H. Porter and E. M. Kesler, *The Pennsylvania State University.*

Forty-eight Holstein calves were used to evaluate high quality legume-grass silage in the diet of the calf from birth. Silage was fed both as the sole source of roughage and as a supplement to hay at two concentrate levels. The calves were randomly assigned to six groups and fed for 16 weeks. All calves received whole milk for six weeks at the rate of 9% of the initial liveweight. Roughage and concentrate were fed daily as follows: Group I (control), starter 5 lb. maximum, hay *ad lib.*; Group II, starter 5 lb. maximum, grass silage *ad lib.*; Group

III, starter 5 lb. maximum, hay and grass silage *ad lib*; Group IV, starter 2 lb. maximum, hay *ad lib.*; Group V, starter 2 lb. maximum grass silage *ad lib.*; Group VI, starter 2 lb. maximum hay and grass silage *ad lib*. The hay and silage contained approximately 85% alfalfa and 15% grasses. The silage fed throughout the trial averaged 28% D.M. Liveweight data showed that within concentrate levels, the treatments were not significantly different. However, the calves on the 5 lb. maximum daily starter intake grew at a significantly greater rate (P less than 0.01) than those on the 2 lb. concentrate level. Roughage consumption was markedly greater for the low level concentrate groups, but was not pronounced enough to overcome the nutrient differential caused by the varied concentrate intake. Differences among treatments were slight with regard to height at withers, and barrel and chest circumference. This trial has indicated that good grass silage can be used as the sole source of roughage for young calves.

LIMITED VS. LIBERAL GRAIN WITH ADEQUATE ROUGHAGE IN SUCCESSIVE LACTATIONS. A. D. Pratt, *Ohio Agricultural Experiment Station.*

A county experimental farm herd of 22 Jerseys was divided to form two comparable groups. Both groups were fed 24 lb. per cow of the same silage and as much hay as they would consume to balance the energy requirement. One group was fed liberally of grain and the other sparingly. In summer both groups grazed together. The amount of grain was the only variable. Both groups contained cows varying in age from 2 to 10 years. The average 305 day production of 4% milk converted to a mature equivalent was greater from the group fed grain liberally. However, those that completed second lactations showed significant trends; those that were fed grain liberally decreased in the second lactation while those fed grain sparingly increased to produce more than the liberally fed group. The trends have continued for those that have completed three lactations.

CHEMICAL COMPOSITION AND YIELD OF FORAGES FERTILIZED WITH 0 TO 400 POUNDS OF NITROGEN PER ACRE. Carroll H. Ramage, Claude Eby, Robert E. Mather, Ernest R. Purvis, Bruce R. Poulton and Frank Wright, *New Jersey Agricultural Experiment Station.*

The difficulty of maintaining satisfactory alfalfa stands on much of the three and one-half million acres of Duchess soil in the Northeast led to the study of the maximum response of grasses to heavy nitrogen fertilization and of alfalfa-grass mixtures to heavy nitrogen and potash fertilization. Four forages were used in the experiment: Orchardgrass, reed canarygrass, alfalfa-orchardgrass, and alfalfa-bromegrass. Fertilizer applications on the grasses were 50, 100, 200, and 400 lb. of nitrogen per acre. On the alfalfa-grass mixtures, three rates of nitrogen (0, 75, and 150 lb.) and two rates of potash (150 and 300 lb.) were used. Dry matter yields the first year for the orchardgrass were (three cuttings) 6423, 6858, 7962, and 9139 lb. per acre respectively. Reed canarygrass yields were 6199, 6722, 7810, and 9217 lb. The alfalfa-orchardgrass yields for the three nitrogen levels were 6008, 5551, and 5950 lb. per acre. For the two potash levels the yields were 5896 and 5829 lb. per acre. Alfalfa-bromegrass yields for the three nitrogen levels were 6424, 7237, and 6941 lb., and for the potash levels 6711 and 7023 lb. Proximate analyses on the grasses showed marked differences in protein content, from 12% to 19%. There was very little difference in any of the components of the alfalfa-grass mixtures. All samples were about 16.5% protein. First cutting yields the second year on low nitrogen grass plots were less than half that of first cutting the first year.

EFFECT OF THYROPROTEIN AND SUPPLEMENTAL FEED DURING THE LATE SUMMER ON THE PERFORMANCE OF BEEF COWS. J. D. Shroder, L. S. Pope, R. W. MacVicar and Dwight Stephens, *Oklahoma Agricultural Experiment Station.*

A four-year study was conducted with 56 Hereford cows to determine the effect of supplemental feed, with and without thyroprotein, on weaning weights of the calves and subsequent reproductive performance of the cows. In four trials, beef cows grazing bluestem pastures and supplemented from July 1 to weaning (mid-October) with 3.5 lb. of oats and 1.5 lb. of cottonseed meal per head daily, weaned calves only 13 lb. heavier than their unsupplemented controls—although the supplemented cows gained 32 lb. more during this period. There was no apparent benefit from supplemental feed during the first year of the trial while the heifers were two-year-olds nursing their first calves.

In another phase of the study, feeding 5.0 lb. of a pelleted supplement containing protamone (1.0 gm. per 100 lb. body weight in two trials, and 1.5 gm. in one trial) increased average weaning weight of the calves by only 14 lb. over the unsupplemented group. A sharp weight loss occurred during the first 28 days when the cows were fed pellets containing the 1.5 gm. level, however there was no apparent effect on subsequent reproductive performance. These studies would indicate that beef cows on good pasture are inefficient in using additional T.D.N. from supplemental feed to increase milk production, and that thyroprotein is of little value in supplements for beef cows nursing calves.

PROTEIN REQUIREMENTS OF GROWING-FINISHING SWINE ON LEGUME PASTURE. V. C. Speer, D. V. Catron, G. C. Ashton and C. C. Culbertson, *Iowa State College.*

Two experiments were conducted, during successive pasture seasons (1953 and 1954), in which 36 cross bred pigs were individually self-fed in each trial from weaning to 200 lb. using individual pig feeding portable pasture units. Allotment was based on weight, sex and breeding. Three pigs in each trial were individually fed a ration containing 8, 10, 12, 14, 16 and 18% protein with and without chlortetracycline. The rations contained ground yellow corn and soybean oil meal, fortified with minerals and vitamins. Pasture units were moved three times weekly. Back fat was estimated by live probe at 200 lb. Daily gains and feed efficiencies to 200 lb. in the first trial were 1.32, 1.58, 1.67, 1.61 and 1.58 lb. per day and 3.42, 3.15, 3.05, 3.04 and 3.24 lb. of feed per pound gain respectively for the 10, 12, 14, 16 and 18% protein levels. Pigs fed the 8% protein ration were terminated at 75 lb. because of poor performance, 0.36 lb. daily gain on 5.32 lb. of feed per pound gain. There was a significant linear and quadratic effect of protein level on rate of gain and feed efficiency. Antibiotic feeding did not effect gains; however, it significantly improved feed efficiency. No significant differences were found in back fat measurements due to ration treatment. Pigs fed the lower levels of protein consumed more pasture than pigs fed the higher levels. Ten week data on the second experiment showed trends similar to the first experiment.

ASSOCIATION OF CHANGES IN RUMEN FILL WITH LIVE WEIGHT OF LACTATING DAIRY COWS. W. E. Thomas, R. D. Mochrie, J. L. Cason and R. K. Waugh, *North Carolina State College.*

Two rumen fistulated cows, a 658 lb. Jersey (J) and an 821 lb. Ayrshire (A) were used in preliminary studies to determine the amount of fluctuation in live wt. that might be accounted for by changes in rumen contents (RC). On 3 con-

secutive days at 2 wk. intervals the following observations were made: total live wt. (TW), wt. of front feet when full, wt. of (RC), wt. of cow minus (RC), and wt. of front feet when empty. The (RC) were mixed and samples taken for dry matter determinations before being returned to the rumen. The kind and amount of concentrate was constant and a different source of roughage was provided *ad libitum* for each 8 wk. period. When fed hay the day to day variation in (RC) accounted for 75% of var. in (TW) for (J) and 84% for (A) and, the between period var., for 99% for (J) and 95% for (A). Similar day to day values on pasture were 44% for (J) and 82% for (A) whereas no period association was observed. Water of (RC) was as highly associated with (TW) as total (RC). Less fill on pasture was evident from the av. wt. and var. of (RC) which were for (J) 96 ± 18 on hay and 90 ± 7 on pasture and for (A) 161 ± 20 and 149 ± 12 on hay and pasture respectively. Most of the var. in (TW) and (RC) was attributable to cows, periods, and days with little due to interaction sources. The change in % (TW) on front feet for full and empty cows could not be used to estimate wt. of (RC). The % (TW) on front feet on hay was for (J) 53.6 full and 53.5 empty and for (A) 52.2 full and 53.4 empty. Similarly on pasture the values were 54.9 when full and 54.7 when empty for (J) and 52.2 full and 53.3 empty for (A).

A MODIFIED CRYOVAC PROCESS FOR MAKING GRASS SILAGE IN THE LABORATORY. B. J. Walker, D. L. Hill and N. S. Lundquist, *Purdue University*.

A modification of the Cryovac process of packaging was used to make grass silage in the laboratory. The process consisted of placing 3 to 5 lb. of fresh chopped forage into a saran plastic bag, vacuumizing (15" Hg, gauge) to remove air and sealing with aluminum clamps. The bags were then stored in a metal cabinet at room temperature (70–85° F.). The pH, odor and appearance of these silages indicated that a typical silage fermentation had occurred. No mold growth was observed in any of the silages.

POLYUNSATURATED FATTY ACID CONTENT OF LEGUME-GRASS SILAGES. Ruth M. Ward, R. S. Allen and N. L. Jacobson, *Iowa State College*.

First cutting non-wilted, chopped legume-grass forage (mostly alfalfa and brome grass) was ensiled in miniature laboratory silos (1 gallon jars), in concrete miniature silos (2 feet in diameter and 6 feet high) and in a stack silo (24×100 feet). Second cutting material was ensiled only in concrete miniature silos. Preservatives included in this study were ground ear corn (stack silo); sodium nitrite, calcium acetate and dried beet pulp (lab. silos); and sodium metabisulfite, calcium formate, ammonium acetate, dried beet pulp and cane sugar (concrete miniature silos). Silage without added preservative also was made in each type of silo. Chemical analyses for linoleic acid, linolenic acid and total long-chain fatty acids were made on the forage as ensiled and on the silages. On a dry matter basis the average percent linoleic acid, linolenic acid and total long-chain fatty acid contents, respectively, were: 0.28, 0.75 and 2.0 (crop as ensiled, 12 samples); 0.33, 0.82 and 2.3 (silages with preservative, 55 samples); and 0.26, 0.87 and 2.5 (silages without preservative, 23 samples). The second cutting material (as ensiled and as silages) tended to be somewhat higher in linolenic acid and total long-chain fatty acids. Stack silage with ground ear corn as a preservative contained more linoleic acid than stack silage with no preservative. None of the preservatives tested in the laboratory or concrete miniature silos had a marked effect on the lipids under investigation.

ALFALFA SILAGE PRESERVED BY VARIOUS METHODS FOR PREGNANT-LACTATING EWES. Patch G. Woolfolk, C. M. Thompson and R. B. Grainger, *University of Kentucky*.

Four lots of 15 pregnant Hampshire cross western ewes each were fed in drylot from December 18 to April 21 (124 days). Lot 1 ewes were fed an average daily ration of 4.0 lb. of U. S. No. 1 alfalfa hay and 0.79 lb. of U. S. No. 2 shelled yellow corn. Ewes in lots 2, 3, and 4 were fed an average daily ration of 1.13 lb. of the same hay, 0.79 lb. of corn plus 8.54 lb. of either molasses preserved alfalfa silage (lot 2), sulfur dioxide-alfalfa silage (lot 3) or sodium bisulfite-alfalfa silage (lot 4). Fleece weights averaged 9.2, 8.2, 8.3, and 8.8 lb. per ewe and bodyweight losses, 8, 14, 15, and 7 lb. per ewe, respectively, for the four lots. At 28 days of age single lambs averaged 24.9, 22.5, 22.0, and 26.0 lb.; twins, 19.6, 14.5, 13.9, and 12.4 lb., respectively, for the four lots. At 56 days single lambs averaged 40.3, 34.9, 36.3, and 40.8; twins, 29.2, 20.4, (none in lot 3), and 20.9. In digestion trials with wethers, apparent digestion coefficients for the molasses, sulfur dioxide, and sodium bisulfite silages were found as follows: Dry matter, 61.0, 61.2, 66.8; crude protein 75.0, 78.9, 79.6; ether extract, 60.1, 53.1, 58.5; crude fiber, 44.9, 43.0, 54.0; N-free extract, 72.1, 71.0, 76.2; energy, 60.4, 60.5, 67.0. Differences in digestibility in favor of sodium bisulfite silage over molasses and sulfur dioxide silages for dry matter, crude protein, crude fiber, N-free extract, and energy were highly significant in all cases except in the comparison of crude protein digestibility with sulfur dioxide silage. Nitrogen retention was 10.41, 11.32, and 10.75 gm. per day, respectively.

Physiology Section

N. L. VanDemark, *Chairman*

EFFECT OF FREQUENCY OF EJACULATION ON THE SEMEN PRODUCTION AND SEXUAL ACTIVITY OF YOUNG DAIRY BULLS. J. O. Almquist, E. B. Hale and R. G. Saacks, *The Pennsylvania State University*.

Nine 2-yr.-old bulls, including two sets of identical twins and one set of identical triplets, were ejaculated either two (control) or six times per week except that one triplet was ejaculated four times per week. After 24 weeks the control and experimental treatments were reversed. Bulls were prepared by 2 minutes of restraint and a false mount. Increasing frequency of semen collection from two to six ejaculates increased the total number of motile spermatozoa produced per week from 1.7 to as much as 3 times. Based on the number of motile sperm per ejaculate, the average output was not altered by four or six ejaculations in three of the seven comparisons. In the remaining four comparisons, six ejaculations per week resulted in decreases of 16, 18, 41, and 43% in the number of motile sperm per ejaculate. No consistent changes in semen characteristics were noted, although two bulls showed a reduction in volume per ejaculate and two in number of spermatozoa per ml. In all instances the required number of ejaculations was obtained. With four and six ejaculations per week, five of the seven bulls required a change of teasers in order to keep the reaction time less than 15 minutes (including coaxing after the first 10 minutes). Presentation of two adjacent teasers simultaneously was effective in reducing reaction time in four bulls not responding to single teasers within 15 minutes. With two ejaculations per week, only two of seven bulls required a change of teasers. The semen and behavior characteristics

noted above for the more frequent ejaculations reverted to pre-experimental values when the bulls were returned to two ejaculations per week.

A NEW INDEX FOR MEASURING THE ADAPTABILITY OF CATTLE TO TROPICAL CONDITIONS. Manuel V. Benezra R., *Central University of Venezuela.*

The Rhoad index as a guide for adaptability of cattle to tropical conditions depends on differences in rectal temperatures. When such body temperatures are the same among a group, those individuals are preferred that under identical conditions show the slowest rate of respiration. With a measurable difference in rectal temperatures the animal with the lower temperature is considered better adapted even if it needed to do more work of respiring in order to maintain its body temperature as near as normal as possible. Rectal temperature used as the only criterion is not a sufficiently sensitive or accurate guide for selecting individuals kept under conditions of shade (open barns or sheds) at least during the hottest part of each day as is the case with a majority of dairy cattle of Venezuela. A formula is proposed differing from the Rhoad coefficient in that it gives definite recognition to respiration as an effort to keep the body temperature normal. It consists of two parts that when integrated, constitute a new coefficient of adaptability B.T./38.33 N.R./23, when B.T. is the body temperature taken in the rectum in Centigrade degrees, 38.33 is the normal body temperature of cattle under most favorable conditions, N.R. is the number of respirations per minute observed at the flank of the animal and 23 is the mean number of respirations per minute under near ideal conditions. The sum of "2" would therefore represent a very high degree of adaptability. Under similar conditions an increase in the figure would mean a lower degree of adaptability. A supplementary index is N.I.R./T.I.T., referring to the relationship of the number of increased respirations per minute for every tenth degree increase in body temperatures.

THE STIMULATORY EFFECT OF CERTAIN FATTY ACIDS ON *IN VITRO* CELLULOSE DIGESTION BY RUMEN MICROORGANISMS. Orville G. Bentley, Alfred Lehmkuhl, Ronald R. Johnson and A. L. Moxon, *Ohio Agricultural Experiment Station.*

Rumen microorganisms have been shown to require unidentified nutritional factors which are needed for rapid cellulose digestion *in vitro*. The addition of rumen juice to a purified medium which was inoculated with rumen organisms separated from strained rumen juice by high speed centrifugation, increased cellulose digestion 3 to 5 fold. When the volatile components were distilled from acidified rumen juice and the distillate added to the *in vitro* fermentation flasks, an increase in cellulose digestion equal to the stimulation observed with the original rumen juice was obtained. However, the distillate from alkaline rumen juice did not affect cellulose digestion—the activity was found to remain in the alkaline residue. This suggested that a volatile substance from rumen juice which behaved chemically like an acid possessed cellulolytic activity for rumen microorganisms *in vitro*. Valeric acid was found to be as active as the distillate from the acidified rumen juice. Isōbutyric, iso-valeric and caproic acids also showed some activity. Water extracts of yeast, dried distillers' solubles, and alfalfa meal were active but this activity could not be distilled off when made either acidic or alkaline. Possibly the activity in these materials is due to their amino acid content. A mixture of pure amino acids stimulated cellulose digestion and several amino acids were found to have some activity; however, of the amino acids tested, valine was the most active. The relationship of these compounds to the cellulolytic factor(s) as found in natural feedstuffs is indicated.

THE FERTILITY OF FROZEN BOVINE SPERMATOZOA AFTER 7-DAYS STORAGE AT — 79° C. R. W. Bratton, Joan C. Cruthers, S. Wearden, R. H. Foote and H. O. Dunn, *Cornell University, New York Artificial Breeders' Cooperative, Inc.*

Portions of 75 first and second ejaculates from 8 top ranking AB proved Holstein sires in the stud of the New York Artificial Breeders' Coop., Inc. were frozen in an extender containing 20% fresh egg yolk, 2.32% sodium citrate dihydrate, 7% glycerol and 500 units each of penicillin and streptomycin. The remaining portion of each ejaculate, preserved unfrozen in an extender containing 50% yolk, 1.45% sodium citrate dihydrate and 500 units each of penicillin and streptomycin, served as a control. The frozen samples were stored at the Central in — 79° C. alcohol using dry ice as the coolant and transported to 149 technicians in the field in specially designed cases under the same conditions. They were thawed on the farm in 30° C. to 37° C. water just prior to insemination. The unfrozen control semen was kept at 5° C. and shipped to technicians in the routine manner. Both the frozen and unfrozen semen were extended at a rate estimated to give approximately 10×10^6 motile spermatozoa per insemination. From motility examinations on the days the semen was used the average number of motile spermatozoa per insemination was estimated to be 7.6×10^6 for the frozen and 8.6×10^6 for the unfrozen semen. The 60- to 90-day percent nonreturns were 73.2 for 1151 first services to the frozen and 71.0 for 1278 first services to unfrozen semen.

FACTORS ASSOCIATED WITH LOW FERTILITY IN BEEF CATTLE. W. C. Burns, A. C. Warnick, M. Koger and A. M. Pearson, *Florida Agricultural Experiment Station.*

Forty-four cows that had failed to calve either one or two years were assembled and bred to a fertile bull at their first estrus. One-half of the cows were killed 3 days after breeding, and the others were allowed to go 34 days after breeding before slaughter. Ovulation rate, fertilization rate, embryonic survival and genital abnormalities were studied. The interval from first exposure to the bull until estrus was 45.1 days for 22 cows with Brahman breeding (group 1) and 12.4 days for 22 cows with English breeding (group 2). Nine percent of the cows in group 1 did not show estrus while 23% did not ovulate although estrus was manifested, and 5% had unilateral pyosalpinx. Forty-five percent of the cows in group 2 had varying amounts of fluid in the uterus, but apparently this fluid did not interfere with fertilization, although embryonic mortality was high in these cows. Pyometra accompanied by a retained corpus luteum occurred in 5% of the cows in group 2. Forty percent of the cows in group 1 had normal embryos at 3 days whereas only 33% had normal embryos at 34 days giving an embryonic death loss of 18%. Sixty-two percent of the cows in group 2 had normal embryos at 3 days whereas none had normal embryos at 34 days giving an embryonic death loss of 100%. Eighteen percent of the 34 day cows in group 1 and 67% in group 2 returned in heat and were added to the 3 day group.

EFFECT OF GENISTIN ON GROWTH AND REPRODUCTION OF THE MOUSE. M. W. Carter, W. W. G. Smart, Jr. and Gennard Matrone, *North Carolina Agricultural Experiment Station.*

Three diets were used in this study. Each contained 80% soybean oil meal, 10% cerelose, 6% Wesson oil, 4% minerals, and vitamins. Dietary differences were as follows: The soybean oil meal used in Diet I (control) was meal from which genistin had been extracted; in Diet II similar meal was used and 2 gm. of genistin

were added per kilogram of diet; the soybean oil meal used in Diet III was unextracted (naturally containing approximately 0.1% genistin). The growth rate as evidenced by weight gains appeared to be the same for each dietary group. The mice on both diets containing genistin (Diets II and III) gave birth to fewer young than those on the diet containing no genistin (Diet I). In number of young born the differences, which were due to a decrease in the number of litters, between the mice on Diet II (added genistin) and Diet I (control) was significant but was not significant between Diet III and Diet I. Neither the number of mice per litter nor the average weight per mouse per litter was affected.

IN VITRO METABOLISM OF DIETHYLSTILBESTROL IN THE BOVINE RUMEN. Edmund W. Cheng and Wise Burroughs, *Iowa State College.*

Diethylstilbestrol when orally administered has been shown to stimulate rate of gain and economy of feed conversion in growing and fattening beef cattle. Information was sought regarding the metabolism of this compound in the rumen and thus the more likely forms of this material that entered the blood stream. Therefore, a chemical method for the quantitative determination of diethylstilbestrol was first developed to ascertain the stability of the compound in the rumen. It was found that when diethylstilbestrol was incubated aerobically with rumen fluid for 24 hours at 40° C., a considerable amount of this hormone substance was destroyed. No appreciable loss was found, however, under anaerobic incubation. The destructive action of the rumen fluid on this compound was inhibited by the heating of the rumen fluid or by the addition of cyanide. This aerobic destruction of diethylstilbestrol appears to be an enzymatic one, probably involving the polyphenol oxidases that may be present in the feeds ingested by ruminants. Since the rumen is primarily anaerobic, it appears likely that no appreciable diethylstilbestrol is lost or transformed in the forestomach when this hormone substance is fed to cattle or sheep.

ESTIMATING THE THYROTROPIN AND THYROXINE SECRETION RATES OF CATTLE. W. W. Crenshaw and C. W. Turner, *Missouri Agricultural Experiment Station.*

Dwarf beef cattle show many of the symptoms of hypothyroidism. Experiments were initiated to determine whether the secretion of pituitary thyrotropin or thyroxine might be asbent or subnormal. I^{131} was used as a tracer. Following the intravenous injection of I^{131} the uptake by the thyroid glands was followed. The count over the thyroid glands increased for approximately 48 hours, then gradually declined. That I^{131} was taken up by the thyroid glands indicated that dwarfs have the capacity of collecting iodine. It is known that exogenous thyroxine depresses the secretion of thyrotropin. When thyroprotein was fed or thyroxine injected preceding the injection of I^{131}, the collection of iodine by the thyroid glands was greatly reduced below normal. Upon the injection of graded doses of thyrotropin (Armours Lab.), to dwarfs continuously fed thyroprotein, the uptake of I^{131} increased in relation to the dosage of thyrotropin. These studies showed that the dwarf cattle pituitary secretes thyrotropin, that the secretion can be inhibited by exogenous thyroxine, and that replacement thereby restores the iodine collecting capacity of the thyroid gland.

THE EFFECTS OF HORMONE INJECTIONS ON BULLS WITH IMPAIRED BREEDING EFFICIENCY. P. T. Cupps, R. C. Laben and S. W. Mead, *University of California.*

Injections of cortisone and hydrocortisone increased the concentration and decreased the percentage of abnormal forms of spermatozoa of two bulls with poor

quality semen. Hydrocortisone was effective at a lower dosage level than cortisone. DOCA was used in one bull, but it had no effect on the semen. Six bulls have been treated with testosterone propionate and long-acting esters of testosterone. Of the six bulls treated with testosterone, only one responded as measured by an increase in libido and a rise in seminal fructose. During the course of injections, two of the six bulls developed small nodules on the penis which disappeared spontaneously after the testosterone therapy was withdrawn. Motility of the spermatozoa has not been changed by any of the hormones used in the present study.

THE GLUCOSE TOLERANCE TEST IN SWINE AND ITS IMPLICATIONS. Irby Bunding, M. E. Davenport and M. A. Schooley, *The Armour Laboratories.*

A limited number of swine were studied, and a significant number of animals showed a "diabetic type" tolerance to a test load of glucose.

RELATIONSHIP BETWEEN WEIGHT AND SEVEN OTHER BODY MEASUREMENTS OF HOLSTEIN FEMALES, AT BIRTH, 6 MONTHS, 12 MONTHS, 18 MONTHS, AND 24 MONTHS. H. P. Davis, *University of Nebraska.*

Holstein females born at the University of Nebraska were measured monthly from birth to disposal from 1922–1942. In this study the numbers varied from 150–158. Close to the Missouri standard at birth, later the measurements were greater, especially in weight. For the various ages, at six month intervals the correlations with weight are as follows: length of top line = .616; .663; .396; .535; .522; length of rump = .660; .763; .495; .574; .538; heart girth = .640; .854; .776; .766; .726; height at withers = .694; .778; .701; .724; .650; height at hooks = .671; .763; .682; .685; .559; depth of chest = .530; .814; .660; .705; .678; width at hooks = .678; .855; .729; .674; .640. All the correlations were significant at the 1% level. No explanation is offered for the decided drop in the correlation for top line with weight, and length of rump with weight at 12 months. The regressions of the several measurements on weight (one centimeter change equals pounds of weight) by periods are as follows: length of top line = 2.48; 10.15; 6.83; 10.45; 10.78; length of rump = 5.52; 20.67; 13.67; 20.59; 20.66; heart girth = 1.56; 6.15; 6.97; 10.19; 10.98; height at withers = 2.85; 9.86; 14.28; 17.69; 17.76; height at hooks = 2.61; 10.51; 12.51; 16.60; 14.18; depth of chest = 2.59; 17.19; 19.05; 29.77; 33.55; width at hooks = 7.25; 21.61; 21.29; 23.90; 26.70.

THE EFFECT OF BOVINE AND PORCINE SOMATOTROPIN UPON LACTATING EWES. Arthur E. Dracy and R. M. Jordan, *South Dakota State College.*

Pretreatment milk yields of 12 lactating ewes were obtained by separating the ewes from their lambs four hours prior to milking. Oxytocin was administered 1–2 minutes before milking to cause milk letdown and ½ of the udder on each ewe was milked once daily for three consecutive days. The ewes were allotted into two groups. Group 1 received intermuscular injections of 25 mg. of bovine somatotropin suspended in 5 cc of saline solution for five consecutive days. Group 2 received a similar type of injection containing porcine somatotropin during the same period. Milk yield per ewe was gathered on both groups as previously described on the 4th through the 6th day after commencing the treatment (treatment period) and 5th and 7th days after discontinuing treatment (post-treatment period). The response to treatment was measured as the change in milk yield using the pre-injection period for comparison. Average milk yields per milking

for the pretreatment, treatment, and post-treatment periods were: Group 1—180.8, 226.3, and 185.7 ml. respectively; Group 2—179.7, 206.8 and 132.0 ml. respectively. Ewes treated with bovine somatotropin increase their milk production 25% during the treatment period. Ewes treated with porcine somatotropin increased their milk yield 15%. Bovine somatotropin exerted a longer lasting effect than the porcine somatotropin used in this experiment.

ENVIRONMENTAL TEMPERATURE AND FERTILITY OF SOUTHDOWN RAMS EARLY IN THE BREEDING SEASON. R. H. Dutt and E. C. Simpson, *University of Kentucky.*

Semen quality has been improved in Southdown rams during the summer months by keeping them in an air-conditioned room at 45°–48° F. In order to compare fertility of rams kept at this temperature with that of control rams, 120 crossbred ewes were bred and the fertility rate, embryonic mortality and lambing percentage were studied. During the last week of May six rams were randomly divided into a treated group of three rams, which was kept in the air-conditioned room, and a control group of three rams, which was kept in the open barn. The ewes were paired as they came into heat, and alternate pairs were inseminated artificially with a split portion of semen from either a treated or a control ram. Breeding was commenced on August 20, and each ram was bred to twenty ewes. One ewe from each pair was slaughtered three days after breeding to determine the fertility rate; the remaining sixty ewes were checked for return to heat and allowed to go to lambing time to furnish information on embryonic mortality and lambing percentage. Fertilization rates were 26.0% for the ewes bred to the control rams and 64.2% for the ewes bred to the treated rams; lambing percentages were 14.3% and 50.0% respectively. Estimates of embryonic mortality were 69.2% for the ewes bred to control rams and 41.2% for the ewes bred to treated rams. Fertility of the rams kept at the lower environmental temperature was significantly higher.

ATTEMPTS AT NON-SURGICAL TRANSFER OF BOVINE OVA. P. J. Dziuk and W. E. Petersen, *University of Minnesota.*

Normal cycling heifers and cows, some of which had produced one or more calves were used as the experimental animals. The animals chosen as the donors were superovulated. They were injected subcutaneously with pregnant mare serum for five days starting on the fifteenth day of the cycle followed by an intravenous injection of an unfractionated sheep pituitary extract (Searles) or with a purified follicle stimulating hormone (FSH) (Armour) followed by a dose of a purified luteinizing hormone preparation (LH) (Armour) or the unfractionated extract, following the injection procedure above. Five days after the intravenous injection, attempts were made to flush the ova from the uterus. A specially constructed self-retaining catheter with ten holes throughout the length of that section introduced into the uterus was used in this procedure. About 200 ml. of homologous serum was used as the flushing medium. When necessary, the recipients were recycled by daily doses of progesterone. A total of 17 superovulations has been attempted. Pregnant mare serum proved to be very inconsistent in producing follicles and the cows seem to become refractory and do not respond to repeated treatments. The purified FSH consistently produced four to six follicles per ovary, but ovulation was not achieved with the purified LH. Ova were recovered in five of 13 attempts with numbers of 1, 1, 3, 5 and 5 ova per flushing and only in those cases in which the follicles were ruptured manually or with the unfractionated pituitary extract. Eight ova that appeared to be dividing normally were transferred to suitable recipients. No pregnancies resulted.

INFLUENCE OF LEVELS AND COMBINATIONS OF ANTIBIOTICS IN BULL SEMEN DILUTED WITH HEATED HOMOGENIZED MILK ON NONRETURN AND NONRETURN DECLINE. R. E. Erb, L. E. Mikota, F. H. Flerchinger and M. H. Ehlers, *State College of Washington.*

Seven split sample trials, each of two months duration, were conducted which involved over 67,000 first and second service inseminations. Semen was diluted with heated homogenized milk. Penicillin (pen.), streptomycin (strep.) and sulfanilamide (sulfa.) were used singly and in combination. Pen. (1000 units/ml.) failed to increase 120-day nonreturns (NR) but did decrease the 29 to 120-day nonreturn decline (NRD) 1.1% ($P < 0.19$) as compared with no pen. Pen. reduced NR 7.3% in semen used the third-day after collection ($P < 0.03$) with no difference between one and two-day old semen. Strep. (1000 mcg./ml.) treated semen NR was 5.4% higher for three-day old semen as compared with untreated (not significant); NR of one and two-day old semen was also slightly improved. Pen. and strep. (1000 units and mcg., respectively) also showed slight improvement with no clear cut evidence of pen. toxicity. Strep. was slightly superior when compared with pen. This was most noticeable for three-day old semen. Pen.-strep.-sulfa. (latter 0.3%/ml.) showed no advantage over pen. and strep. Strep. (1000 mcg./ml.) was slightly superior to 500 mcg./ml. of strep. and 500 mcg. and 500 units of strep. and pen., respectively. Use of antibiotics singly and in combination effectively eliminated between bull differences in NRD. Strep. (1000 mcg./ml.) was equal to or superior to pen. and sulfa. singly and/or in combination with strep. for increasing NR and decreasing NRD.

THE PHYSIOLOGY OF DWARFISM IN BEEF CATTLE. J. M. Fransen and F. N. Andrews, *Purdue University.*

Detailed physiological studies have been conducted on sixty-six dwarf calves. Gross studies of 44 dwarf thyroids have revealed an average thyroid weight of 10.96 gm. Twenty-six calves had glands below and 18 above the average thyroid weight. Nine animals with cystic pituitaries and 21 animals with cystic adrenals were observed at autopsy. In some instances the same animal had both cystic adrenals and a cystic pituitary, and in three instances bilateral adrenal cysts were observed. Reproductive tracts and gonads of both sexes have been within the normal range. In a large percentage of cases internal hydrocephalus with distention of the lateral ventricles was observed. Grossly, the remainder of the glands and organs have been relatively normal in most instances. Histological studies have also revealed a wide range of activity in most glands and organs; however, most have been within a normal range. Blood cholesterol values have varied greatly within the same animal and between different animals; however, in most instances it has been within the normal range. Bioassay of dwarf pituitaries has shown normal thyrotropic and gonadotropic hormone activity. Cerebrospinal fluid pressure has been high in all cases. Electrocardiographic studies revealed normal heart activity in nine of ten dwarf animals. Hormone therapy of 11 dwarf calves with thyroprotein, stilbestrol and testosterone, singularly or in combination, stimulated average daily gain and general behavior of the treated calves, but did not significantly alter bone growth. Hematology studies, in most instances, have been within the normal range for similar aged beef calves.

THE MAINTENANCE OF PREGNANCY IN THE VITAMIN A DEFICIENT RABBIT WITH PROGESTERONE. R. L. Hays and K. A. Kendall, *University of Illinois.*

Fifty-eight adult New Zeland White does were placed on a diet containing no detectable carotene or vitamin A for varying lengths of time and were then

mated to fertile males. All mated females were autopsied 28 days after mating. In the first experiment the females were mated after 4, 8 and 12 weeks of vitamin A depletion. One-half of the females received daily injections of 8 mg. of progesterone. The most pronounced effect of progesterone was observed in the group depleted for 12 weeks. The 6 progesterone treated females averaged 4.3 living and 2.5 dead young with 9.8 corpora lutea as compared to the untreated females which averaged 0.8 living and 1.2 dead young with 1.0 corpus luteum. The liver vitamin A declined from 10 gamma per gram in the 4 week group to 2 gamma or less in the 12 week group. In the second experiment 24 females were mated after 13 weeks on the vitamin A deficient diet and then divided into 3 groups. The untreated group averaged 1.0 living and 2.6 dead young, with 5.0 corpora lutea. The group receiving 12.5 mg. progesterone daily averaged 5.1 living and 2.6 dead young with 10.25 corpora lutea and the group receiving 100,000 I. U. of vitamin A acetate weekly averaged 1.2 living and 1.8 dead young with 5.5 corpora lutea. The liver stores of vitamin A in the first two groups were below .5 gamma per gram while the latter group averaged 787 gamma.

THE RELATION OF AMBIENT TEMPERATURE TO WEIGHT GAIN IN SWINE. Hubert Heitman, Jr., C. F. Kelly and T. E. Bond, *University of California and U. S. Department of Agriculture.*

The results of six years work on the relationship between constant ambient temperature and gain in weight have been summarized and submitted to statistical analysis. Pigs weighing approximately 50 to 125 lb. showed maximum weight increase at about 73 degrees F. while swine weighing about 150 to 350 lb. showed maximum weight increase at about 61 degrees F. The data in each case fit a second degree curvilinear regression. Confidence limits have been fitted to the data. The data on feed utilization show a similar, but inverse relationship to ambient temperature.

AGE TRENDS IN GONADOTROPHIC HORMONE IN FEMALE SWINE PITUITARIES. R. Hollandbeck, B. Baker, Jr., A. V. Nalbandov and H. W. Norton, *University of Illinois.*

The gonadotrophic hormone content of the pituitaries of 26 Hampshire and 20 Duroc females, varying in age from 1 to 1330 days, including cycling and pregnant animals, was assayed using day-old male chicks. The results were correlated with age and body weight. No significant difference was found between breed means of open sows. The unit potency decreases linearly as age increases ($P < 0.01$). Unit potency was not clearly related to body weight. The breed means of the pregnant sows differed significantly ($P < 0.05$), Hampshires giving 11.6 mg. greater chick testes weight. Total potency for open sows shows no difference in breed means and increases linearly with age ($P < 0.02$). The amount of available hormone per unit of body weight is very high in day old females, decreases rapidly to about 220 days and remains constant until 1330 days of age. The onset of puberty coincides with the lowest titer of available hormone. This apparent drop in available hormone is probably due to a shift in the type of gonadotrophic hormone secreted rather than to a decrease in total hormone available.

FURTHER STUDIES ON FACTORS AFFECTING OVULATION IN THE COW. W. H. Hough, H. J. Bearden and W. Hansel, *Cornell University.*

In previous studies it was found that: (a) atropine given at the beginning of estrus blocked ovulation in dairy heifers. (b) Ovulation occurred at the normal

time or earlier when atropine and luteinizing hormone (chorionic gonadotrophin) were given simultaneously at the beginning of estrus. (c) Progesterone given at the beginning of estrus hastened ovulation and (d) estradiol given at the beginning of estrus did not hasten ovulation. In the first of the present studies the length of estrus and the time of ovulation were compared in a control estrus period and in an estrus period in which epinephrine was administered at the beginning of heat in 14 heifers. Epinephrine administration did not significantly affect ovulation time; the average time elapsing from end of estrus to ovulation was 11 hours in the control periods and 9.8 hours in the treated estrus periods. In a second study the time of ovulation was compared in an estrus period in which atropine was administered at the beginning of heat and in an estrus period in which both atropine and progesterone were administered at the beginning of heat in 11 heifers. Ovulation was blocked in 7 of the 11 heifers when atropine alone was injected and in 4 of the 11 heifers when both atropine and progesterone were given. This difference is not significant when tested by chi square. These data are probably best interpreted as indicating that the ovulation hastening effect of progesterone in normal heifers is mediated through the hypothalamus rather than by a direct action on the anterior pituitary gland.

COTYLEDONARY ATTACHMENTS OF RANGE EWES. Art Hoversland, H. Watling and J. L. Van Horn, *Montana Agricultural Experiment Station.*

A study was initiated to investigate what effect type of birth, age of ewe, size of ewe, birth weight of lambs, and weight gain of ewe during pregnancy had on total number and total area of cotyledonary attachments. The extra-embryonic membranes of 78 range ewes of Rambouillet, Targhee and Columbia breeding were obtained immediately after lambing. The number of attachments and the diameter measurements of at least 10% of the attachments were recorded. These data were taken separately from the chorion extruded from each horn of the uterus. The average number of attachments was 78, ranging from 40 to 124. The mean total calculated area of these cotyledonary attachments was 415 square centimeters with a range from 169 to 1311 square centimeters. In single births the mean number of attachments was 77 and mean area was 330 square centimeters. In multiple births the mean number of attachments was 87 and the mean total area was 462 square centimeters. In single births the pregnant one-half of the chorion had a mean of 45 attachments with a mean total area of 223 square centimeters as compared to the non-pregnant one-half having a mean of 33 attachments and a mean total area of 94 square centimeters. The chorion in single births was one single sac extending into the non-pregnant horn of the uterus with attachments present there. The chorion from multiple births was also one single continuous sac. On several chorions one or two extremely large oblong attachments were noted in the area from the body of the uterus. The cotyledonary attachments on some chorions were uniformly large or small and others had extremely varied sizes present.

THE EFFECTS OF SHEARING AND LEVEL OF FEEDING ON FERTILITY IN SHEEP. C. V. Hulet, A. ElSheikh, A. L. Pope and L. E. Casida, *University of Wisconsin.*

Groups of 20 rams and 80 ewes were each divided equally as lambs into two lots one of which received roughage-plus-grain (a fattening ration) and the other only roughage. All were sheared in April and half of each group of rams was resheared at monthly intervals until breeding was completed. Breeding was from

August 20 to October 20 when the sheep were yearlings. Each ram was mated to two ewes from each lot. One of these ewes from each lot was slaughtered at 3 days and the ova recovered to determine fertilization and the remaining ewe from each lot was slaughtered at 40 days to determine embryo survival. In addition various semen characteristics were studied throughout the breeding period. The sheared rams had a significantly higher fertilization rate than the unsheared rams. Likewise, a significantly larger proportion of ewes bred to the sheared rams had normal embryos at 40 days gestation than those bred to the unsheared rams. No difference in fertilizing ability was found between the rams on the two different levels of feeding. The roughage-plus-grain fed ewes had more normal embryos both at 3 days and 40 days than the other group but the estimated percentage embryo survival was lower in this group than in those receiving only roughage. No significant relationship was found between the various measures of semen quality and fertility though the group with the highest fertility also had the best semen quality.

CAUSES OF BABY PIG MORTALITY. H. D. Hutchinson, S. W. Terrill, C. C. Morrill, H. W. Norton, R. J. Meade, A. H. Jensen and D. E. Becker, *University of Illinois.*

Two years of data from the University of Illinois herd involving 2116 pigs in 220 litters of four breeds in four farrowing seasons are summarized as to causes of deaths before weaning. Of 657 (31%) deaths, 109 (17%) were born dead. 174 (26%) died the first day, 86 (13%) the second, 60 (9%) the third, and 36 (5%) the fourth. Of 548 dying post partum, 125 (23%) died of inanition, 104 (19%) of inanition and congenital weakness, 82 (15%) were overlain, 43 (8%) died of arthritis, 30 (5%) of congenital weakness, 30 (5%) of inanition and being overlain, and the remaining 134 (24%) of miscellaneous causes and combinations including 58 (11%) undiagnosed.

CAROTENE AND VITAMIN A LEVELS IN BOVINE PLASMA AND COLOSTRUM ASSOCIATED WITH PREPARTUM PROGESTERONE ADMINISTRATION. K. A. Kendall, R. L. Hays, L. E. McDonald and G. D. Rolleri, *University of Illinois.*

In connection with another investigation in which 500 milligrams of progesterone were administered to each of 4 purebred dairy cows 33, 23 and 13 days plus 100 milligrams of progesterone given to each cow 3–2 days before parturition, the daily levels of carotene and vitamin A in the plasma and colestrum were compared with the levels of 4 similar control cows without progesterone. Blood samples were taken at the same time each day for a period of 36–40 days before the expected calving date and for 7 days at the 14th and 21st days after parturition. The results suggest that progesterone had little or no effect upon plasma carotene levels; however, the weekly average plasma vitamin A levels appeared to be higher particularly during the first 2 weeks following calving. The average plasma vitamin A levels (micrograms per 100 ml.) for the first 7 days following parturition for the progesterone treated cows were 28.0, 16.5, 31.0 and 25.1 while the plasma values for the nontreated cows during the same period were 14.0, 10.5, 8.4 and 14.0. A similar trend was noted at the 14th day following parturition, however, on the 21st day the trend was reversed. The concentration of vitamin A per lb. of butterfat in the first 8 complete milkings in 3 of the 4 progesterone treated cows was lower than those for the controls.

TEMPERATURE CONDITIONS CRITICAL TO MILK PRODUCTION IN HOLSTEIN CATTLE AT BELTSVILLE. Douglas H. K. Lee, H. W. McMullan, R. E. McDowell and M. H. Fohrman, *Johns Hopkins University and U. S. Department of Agriculture.*

Correlations are reported between the milk production of lactating Holstein cattle and summer weather conditions at Beltsville, Md., after standardization of production figures to a theoretical first day of lactation by the formula: $P = p/(1-.00913d)$, where p is observed production at d days after calving. Correlation with dry and wet bulb temperatures is most marked when they relate to the fourth and fifth preceding days. It is substantially similar with maximum and minimum temperatures. Production in cows kept in open barns declines importantly when local mean shade temperature exceeds 71° F. for two successive days. ($p = 129.8 - .829m$, where p is production in lb./day, and m local mean shade temperature in ° F. on 4th and 5th preceding days.) These effects, noticeable in the herd averages, are obscured when individual records are considered. For the conditions prevailing at Beltsville, climatic protection is suggested when local mean shade temperature on two successive days exceeds 66° F. for cows in open field, or 71° F. for animals in open barns.

SOME FACTORS IN FEED LOT BLOAT. Ivan L. Lindahl and Russell E. Davis, *U. S. Department of Agriculture.*

The following diets were studied in relationship to feed lot bloat: *1A,* 14 lb. of concentrate (78.3% corn, 20.7% soybean oil meal, and 1% salt) and 4 lb. of No. 2 alfalfa hay per animal per day; *1B,* same concentrate as 1A with 4 lb. alfalfa meal replacing the hay; *1C,* same as 1A except the salt contained 1 part of a sulfated mono-glyceride type household detergent to 4 parts of salt; and *2A, 2B,* and *2C,* which were the same as above except that barley was substituted for corn. The experiment was divided into 4 thirty-day periods. During the first 90 days, 3 beef-type animals received each of the corn diets for 30 days in a reversal type test and 3 additional animals received the barley diets in the same manner. During the last 30 days of the experiment all animals that had received the corn diets were put on diet 2B and the animals that had received the barley diets were put on diet 1B. Bloat was obtained on all diets and no significant differences were noted between the diets within the treatments or between diets 1B and 2B in the incidence of bloat. A significant difference was noted between animals in their susceptibility to bloat and a significant increase in the incidence of bloat with time, irrespective of diets, was also noted.

THE PROTECTIVE EFFECT OF IN-VIVO WOOL AGAINST RADIOACTIVE "FALL OUT." C. C. Lushbaugh, J. F. Spalding and D. B. Hale, *Los Alamos Scientific Laboratory.*

Studies were conducted to compare beta ray skin lesions produced on Utah range sheep with lesions found on animals after atomic bomb test operations at the Nevada Proving Grounds. Range type ewes were irradiated with beta radiation from Sr^{90}-Yt^{90}. The radiation was delivered at the surface of the wool and at the surface of the shaved skin. Superficial lesions developed in the shaved areas in one ewe after a minimum exposure of 2500 rep and in a second with a minimum dose of 5000 rep. A lesion developed in one animal when a dose of 145,000 rep was delivered at the surface of the wool. Another animal developed a very superficial lesion with a dose of 45,000 rep. The protective effect of the wool was due,

primarily, to the length of the fleece, which decreased the radiation dose to the skin surface in keeping with the inverse square law. The density of the wool also had some protective effect. With a staple length of 33mm and a calculated density of 46.2 mg/cm^3 only 3% of the beta radiation delivered at the surface of the wool reached the skin surface. The experimentally produced lesions differed histologically but not grossly from the lesions observed on the Utah range sheep. The minimal amount of beta radiation necessary to cause epilation in sheep was found to be 400–500 rep.

STEROIDS IN LAMB FATTENING. H. G. Luther, C. R. Adams, H. E. Downing, W. M. Reynolds, G. E. Hawley, *Chas. Pfizer & Co., Inc.*

An implant of 250 mg. of progesterone and 10 mg. of estradiol as a growth stimulant in 50 groups of 18 lambs was employed. The variable included energy content of ration, sex, time of implant, grade of animal, weight of animal at time of implant. In this trial, the improvement of rate of gain as result of implant was independent of ration, sex, implant time, weight of animal and its initial live grade. The rate of gain for all implanted groups was .61 lb./day as contrasted to .46 for the control. The response was essentially comparable on both high and medium energy rations. However, improved rate of gain was noted for the higher energy ration and economic analysis indicates this to be the more desirable. The high energy contains 49% corn, 41% hay; the medium energy 60% hay and 31% corn. Five percent linseed meal was employed in both rations. Implantation had a significant effect on feed efficiency, treated animals consuming 6.3 lb. of feed per pound of gain as compared to 8.3 for the control. Improvement in feed efficiency as a result of implants is independent of sex, weight, grade, time of implant, the most economic results being obtained on the high energy rations. Carcass data show a decrease in 1/3 of a standard grade and a 2% reduction in dressing percentage for the implanted group.

STEROIDS IN CATTLE FATTENING. H. G. Luther, C. R. Adams, H. E. Downing, W. M. Reynolds, G. E. Hawley, *Chas. Pfizer & Co., Inc.*

In a trial designed to evaluate oral diethylstilbestrol and implants of progesterone estradiol, progesterone diethylstilbestrol and estradiol-testosterone, 850-pound Hereford steers were employed in replicate groups of 12 animals per pen. Animals were self-fed a mixed ration of 32% chopped alfalfa, 44% yellow corn, 13% oats, 6½% bran, 2% linseed oil meal and 2% soybean, oil meal. Oral diethylstilbestrol at a level of 10 mg per head per day gave a 35% increase in rate of gain, and a feed efficiency of 10.4, as compared to 13 for the control group. All implant groups gave an increase in rate of gain of 42 to 43% over the control and feed efficiency ranged from 9.6 to 10.0. The progesterone was used at a level of 1500 mg in the implant, testosterone being employed at 300 mg. Estradiol and diethylstilbestrol were used at a level of 50 mg in the implant. One group of the animals was maintained on the steroid treatment for 98 days; the other group being maintained on treatment for 126 days.

IN VIVO UTILIZATION OF NON-PROTEIN NITROGEN BY SHEEP. N. D. Magruder and C. B. Knodt, *Pennsylvania State University.*

In vivo studies using three yearling Hampshire wethers as replicates demonstrated protein synthesis when urea, dicyanodiamide, ammoniated hemicellulose and soybean oil meal supplemented rations were compared to a basal control.

THE REACTIONS OF ¾ SINDHI ¼ JERSEY HEIFERS TO HEAT, AS COMPARED WITH THE REACTIONS OF JERSEY AND ½ SINDHI—½ JERSEY HEIFERS. R. E. McDowell, H. W. McMullan, D. H. K. Lee and M. H. Fohrman, *U. S. Department of Agriculture.*

Studies of the comparative values for rectal temperature before exposure, and the mean rectal temperature during exposure to a test atmosphere at 105° F., with a vapor pressure of 34 mmHg (wet bulb 92° F.) have been made for Jerseys, ½ Sindhi–½ Jersey, and ¾ Sindhi–¼ Jersey heifers 6 to 22 months of age. The results indicate the three groups differ very little in initial rectal temperature and there is a slight decline with age in all groups. It is evident that the mean temperature of the ¾ Sindhi animals during exposure closely resembles that of the ½ Sindhis and the responses of the two crossbred groups do not differ significantly from each other; but the responses of both differ from that of the Jersey group to the same highly significant degree. The results for rectal temperature show no greater success on the part of the ¾ Sindhis over the ½ breds in regulating their body temperature but they are able to do so at least up to 16 months of age, with a somewhat lower respiratory effort. However, it is doubtful whether this slight increase in adaptibility to heat is economically worth the drop in size and milk producing capacity which has been observed.

SEASONAL VARIATIONS IN THE BLOOD COMPOSITION OF SHEEP. P. N. Mehrotra, D. N. Mullick and N. D. Kehar, *Indian Veterinary Research Institute.*

It was reported from this laboratory that cattle maintained the body temperature by adjusting the levels of the metabolites in the blood under varying atmospheric conditions. The present investigation was undertaken to record the blood composition of sheep during different seasons of the year. Nine healthy adult male sheep were fed the same maintenance ration for the whole of the experimental period. Whole blood was analysed for RBC, WBC, Hb, CV and sugar and serum was analysed for protein, nonprotein, calcium, inorganic phosphorus and magnesium at monthly intervals for one year. The maximum and minimum values of air temperature were 105 and 65° F. in June and January and of relative humidity were 85 and 34% in August and May respectively. The mean summer and winter values of RBC, Hb, and CV were 8.4, 9.5 millions; 7.0, 8.6 gm. and 34.5, 38.7% respectively. The figures for protein, nonprotein, sugar, calcium, inorganic phosphorus and magnesium in summer and winter months were 6.8, 6.4 gm.; 41.9, 42.0 mg.; 48.0, 45.0 mg.; 11.8, 12.8 mg.; 5.43, 6.30 mg.; and 2.43, 2.36 mg. respectively. These constituents changed significantly with the variations of the atmospheric conditions due to homeostatic mechanism of the animals.

SEASONAL VARIATIONS IN THE FEED AND WATER INTAKE OF SHEEP. P. N. Mehrotra, D. N. Mullick and N. D. Kehar, *Indian Veterinary Research Institute.*

It was shown from this laboratory that cattle reduce the thermogenesis by taking voluntarily lesser quantities of feed under climatic stress. The present observation was designed to record whether sheep followed the same trend of adjustment of the metabolic behaviours of the nutrients like cattle under different climatic conditions. The digestibility trials and water metabolism were conducted on nine healthy adult sheep (70 to 80 lb.) in different seasons of the year. The average values of the temperature and moisture were 103, 87, 68 and 86° F. and 30, 79, 42 and 47% in summer, autumn, winter and spring seasons respectively. The average values of water intake and insensible perspiration were 3.93 and 2.64 kg. and 3.06 and 1.25 kg. in summer and winter months respectively.

The dry matter ingestions were 872 and 996 gm. and the coefficients of digestion were 50 and 53% in two seasons. The intakes of protein, fat and carbohydrate in summer months were 108.4, 32.3, and 671.3 gm. and in winter months were 115.9, 38.6 and 816 gm. respectively. The mean values of the intakes of calcium, phosphorus and nitrogen were 13.03, 7.87 and 17.35 gm. in summer and 23.10, 10.96 and 18.56 gm. in winter month. The intermediate values of these nutrients were recorded during autumn and spring. It was concluded that sheep relieved the discomfort of climatic stress by voluntarily adjusting the ingestion of nutrients.

EFFECTS OF CORTISONE AND GROWTH HORMONE ON INITIATION OF LACTATION IN RABBITS. Joseph Meites and T. M. King, *Michigan State College.*

Several lactational physiologists have expressed the view that the adrenal cortex and growth hormone have essential and probably specific functions in the initiation and maintenance of lactation. In some preliminary experiments, an attempt was made to determine whether or not injections of cortisone and/or growth hormone would enhance the action of prolactin in initiating lactation. Mature female rabbits were ovariectomized and injected for 25 days with a combination of estrone and progesterone which produced excellent mammary growth. At the end of this period, they were divided into four groups and treated for eight days with (1) prolactin only; (2) prolactin and cortisone; (3) prolactin and growth hormone; (4) all three hormones. On the 9th day the rabbits were killed and their mammary glands were exposed and rated visually for lactational response. The following ratings were assigned each group: (1) 10.2 or excellent; (2) 7.3 or moderate inhibition; (3) 2.3 or marked inhibition; (4) 4.6 or intermediate inhibition between groups 2 and 3. Thus cortisone interfered slightly and growth hormone strongly with the initiation of lactation by prolactin.

THE INFLUENCE OF SEX HORMONES ON THE FATTENING OF UTILITY AND CULL SPRING LAMBS. R. L. Murphree, C. L. Cannon, J. A. Odom, and C. S. Hobbs, *University of Tennessee.*

Two trials involving 37 and 51 lambs were conducted to determine the effects of exogenous sex hormones upon the growth performance of utility and cull spring lambs. In the first trial, neither the subcutaneous implantation of (1) 24 mg. of stilbestrol or (2) 24 mg. of stilbestrol and 200 mg. of progesterone exerted any apparent effect upon the rate of gain in lambs during a 49 day pasture period. During an 80-day feeding period, the average daily gains for the control, stilbestrol and stilbestrol-progesterone groups were .29, .42 and .37 lb. respectively. The stilbestrol treated lambs gained at a 44.8% faster rate on 37.5% less feed and the stilbestrol-progesterone group gained 27.6% faster on 29.8% less feed per pound of gain than the control group. The live animal grades, carcass grades and dressing percentage averaged slightly lower for both hormone treated groups than for the control group. In the second trial, the treatments were: (1) control, (2) 12 mg. of stilbestrol and (3) 12 mg. of stilbestrol and 175 mg. of progesterone implanted subcutaneously. The average daily gains for the three groups during a 56 day feeding period were .075, .22 and .24 lb., respectively. The performance of the control group was disappointing, particularly in view of the fact that during a 70 day pretreatment period on pasture the average daily gains for the three groups were .32, .33 and .32 lb., respectively. The average live animal grades for the control, stilbestrol and stilbestrol-progesterone groups were low choice, choice and high good, respectively at the end of the feeding period. No cases of rectal and/or vaginal prolapse were encountered in either trial.

BREED DIFFERENCES IN SEMEN CHARACTERISTICS OF FAT-TAILED SHEEP. M. M. Oloufa, Y. H. Darwish and A. L. Badreldin, *Cairo University.*

Egyptian sheep belong to the fat-tailed type; Rahmani and Ossimi being the two main breeds of sheep widely distributed in the Nile Delta. Three Rahmani and nine Ossimi rams, 1.5 to 2.5 years old, were used in the present experiment. They were from the College flock on which inbreeding was practiced for over 12 years. Semen was collected from each ram once every 10 days by the artificial vagina. During the experimental period of 12 months, a total of 103 and 282 collections were taken from Rahmani and Ossimi rams respectively. Semen was examined for volume, motility, concentration, percentage of abnormal sperm, and percentage of live sperm in fresh semen and in semen stored for 48 hours at 40° F. While volume, concentration, percentage of abnormal sperm and percentage of live sperm in stored semen were higher in Rahmani than in Ossimi, the reverse was true in the case of motility and percentage of live sperm in fresh semen. Among the criteria studied for semen quality, only semen volume, motility and percentage of live sperm in fresh semen showed statistically significant differences between the two breeds. These differences in semen quality between the two breeds may be due to inherited characters controlling the activity of the pituitary gland or the general endocrine system. It is also possible that they are due to differences in body size, in testis size, or in the efficiency of the scrotum in its role as a thermoregulatory mechanism.

EFFECTS OF HOT WEATHER AND HIGH HUMIDITY ON SEMEN PRODUCTION AND FERTILITY OF THE DAIRY BULL. T. E. Patrick, J. E. Johnston, H. C. Kellgren, J. B. Frye, Jr., Gerald D'Arensbourg and Cecil Branton, *Louisiana State University.*

This investigation was initiated in an effort to determine the effects of temperature and humidity upon semen production and fertility of dairy bulls. Two groups, each consisting of four bulls were used in this study. The bulls in group I were kept in a controlled chamber from June through August where the temperature and humidity were maintained at a constant level. The temperature was maintained at approximately 80° F. and the humidity at 19 mm. Hg. Bulls in group II were housed in the barn at the Dairy Improvement Center under natural environmental conditions. Measurements of physiological responses were taken on all eight bulls. However, semen production and fertility records were kept on only two bulls in each group. Ninety-one percent of semen samples collected from the bulls in group I were suitable for use in artificial breeding as compared to only 47% in the control group. Breeding efficiency for the bulls maintained in the cooler temperature was 73.6% whereas the bulls kept under natural environmental conditions had a breeding efficiency of 71.9%. The two bulls in the latter group had very few services in July and August due to the poor quality of their semen. Information obtained from this study indicates that the semen production and fertility of dairy bulls can be maintained at a relatively high level during the summer months, if they are housed in a barn where the temperature is kept at approximately 80° F.

VARIATION IN RECTAL TEMPERATURE, PULSE RATE AND RESPIRATORY RATE OF CATTLE AS RELATED TO VARIATION IN FOUR ENVIRONMENAL VARIABLES. Fazlu Rahim Quazi and Robert R. Shrode, *Agricultural and Mechanical College of Texas.*

A series of four twenty-four hour studies was conducted with ten Jersey females, ten Holstein females and ten ½ Jersey, ½ Brahman females. Thirteen

observations each of the rectal temperature, pulse rate and respiratory rate of each animal were recorded during the twenty-four hour period, *i.e.*, at the beginning of the test period and at intervals of two hours. Continuous records of solar radiation, air temperature, vapor pressure and wind velocity were obtained. The primary objective was not a breed comparison, but the data conform to the findings of previous studies with respect to the relative heat tolerance of the three groups studied, the order of heat tolerance being: crossbreds most heat tolerant, Jerseys intermediate, and Holsteins least heat tolerant. The data were studied by multiple regression methods using as independent variables the four environmental variables and as dependent variables those measured on the cattle. The results indicate that the response of all three groups was appreciably influenced by air temperature and solar radiation. The Jerseys and Holsteins were less influenced by vapor pressure than the crossbreds. Wind velocity showed a stronger relationship with crossbred rectal temperature than with that of Jerseys and Holsteins.

THE USE OF KETOGESTIN AND SODIUM ETHYL OXALACETATE IN THE TREATMENT OF HYPOGLYCEMIA AND KETONEMIA. E. F. Reber, L. R. Bain, R. D. Hatch, C. E. Schoettle and J. Sampson, *University of Illinois.*

Five clinical cases of bovine ketosis were treated with 11-ketoprogesterone (ketogestin) alone or with glucose and/or other substances. One cow, with mild ketosis, showed improvement within 24 hours after receiving four grams of 11-ketoprogesterone and recovered without additional treatment. A more severe case of ketosis was given two grams of 11-ketoprogesterone for each of two treatments. This case slowly improved 72 hours after the first treatment. In three cases of ketosis the 11-ketoprogesterone was of negligible value. Glucose therapy was found to be most effective in overcoming the ketosis of the latter three cows. 11-ketoprogesterone had a negligible effect upon the blood sugar levels of baby pigs fasted for five days. Hypoglycemia and ketonemia were produced in a pregnant ewe by fasting or feeding hay only. The administration of 11-ketoprogesterone to this ewe lowered blood ketone levels and raised blood sugar levels, but not consistently. Sodium ethyl oxalacetate, given orally to this ewe, was effective in reducing the ketonemia and slightly raising the blood sugar levels.

FACTORS INVOLVED IN THE RADIOACTIVE TRACER METHOD FOR DETERMINING THYROID SECRETION RATE AS SHOWN BY STUDIES IN RATS. E. P. Reineke and O. N. Singh, *Michigan State College.*

Adult Albino rats were injected with 30–40 microcuries of I^{131} and thyroidal retention of iodine was determined by external thyroid counts. Daily thyroxine injections in graded doses were started 48 to 72 hours after I^{131} administration. In some experiments the thyroxine dosage was increased progressively every 48 hours in each rat, an external thyroid count being taken immediately before each increase in dosage. I^{131} loss decreased regularly with increasing thyroxine dosage until the estimated thyroxine secretion level was reached or exceeded. The thyroid secretion rate was determined as the minimum dosage of L-thyroxine required to inhibit further output of I^{131} from the thyroid. To obtain a statistically significant correlation between thyroxine dosage and thyroidal I^{131} retention for individual animals 5 to 6 points were required on the ascending portion of the dose-response curve. For comparison by another technic the thyroxine dosage was varied between groups, each group receiving a constant dose of thyroxine for periods up to 7 days. If the thyroid count taken at the end of the dosage period for each group is expressed as the percent of the preinjection count and plotted against thyroxine dose, the thyroid secretion rate can be estimated as before. Results by both pro-

cedures check very closely, ranging from 2.12–2.56 ug. per 100 gm. body weight. I^{131} output was reduced by thyroxine within 24 hours of administration. Thiouracil administered concurrently with thyroxine to block recycling of I^{131} caused an increase of about 10% in the estimated secretion rate.

A METHOD FOR THE EXTRACTION, PURIFICATION, AND CHEMICAL ASSAY OF 17-HYDROXYCORTICOSTEROIDS IN BLOOD OF CATTLE. W. G. Robertson and J. P. Mixner, *New Jersey Agricultural Experiment Station.*

A method is presented for the extraction, purification and chemical assay of free 17-hydroxycorticosteroids in the blood plasma of dairy cattle, together with some physiological data supporting the usefulness of the method. Blood plasma from dairy cows was chilled to 4° C., salted with 10 gram % of sodium chloride, and extracted with cold ethyl acetate in a cold separatory funnel for 6 seconds. The dried residue of the ethyl acetate extract was partitioned between 80% methanol and hexane, the methanol phase was dried down, and the residue was taken up in absolute methanol for assay. The Porter and Silber method for assay of 17-hydroxycorticosteroids (J. Biol. Chem. 185:201, 1950) was adapted to a 1.0 ml. micro reaction. Compound "F" was selected as the standard material and quantities of "F" ranging from 2 to 20 micrograms were assayed with precision using the Beckman Model B Spectrophotometer. On standard curve data the coefficient of variation of the error term was 4.4%, while the standard error of a single determination of Compound "F" was 0.26 micrograms. Normal plasma values for 17-hydroxycorticosteroids ranged from 7.00 to 17.62 micrograms percent with a mean of 11.78 micrograms percent. The intramuscular injection of 600 Armour Veterinary Units of ACTH (gel form) into each of four cows caused an average increase of 98.76% in the plasma corticoid values in a two-hour period. This increase was highly significant statistically ($P < 0.01$).

NEUTRAL 17-KETOSTEROIDS IN THE URINE OF NORMAL AND DIETHYLSTILBESTROL TREATED WETHER LAMBS. W. S. Ruliffson, T. D. Bell, J. S. Hughes, A. Nunn, *Kansas State College.*

In addition to increasing the rate of growth, the implanting of diethylstilbestrol pellets into wether lambs may cause two undesirable effects. Hydration of body tissues may be increased to such an extent as to lower carcass grade. Secondary male tissue (Cowper's gland, prostate, seminal vesicles, etc.) may be enlarged to such an extent as to cause death due to the prevention of urination. This enlargement may be caused by the direct action of diethylstilbestrol. It is more likely that it is caused by male-like hormones (neutral 17-ketosteroids) produced by the adrenal glands, under the influence of ACTH. Diethylstilbestrol probably causes the pituitary gland to produce increased amounts of ACTH. If the growth of the male tissue is caused by male-like hormones produced by the adrenal cortex, there should be an increased amount of these compounds in the urine. To secure information concerning this two 24-hour samples of urine were collected from two treated and two untreated wether lambs at the conclusion of 120-day feeding test in the Spring of 1953. Three additional samples were collected from each of three treated and untreated animals from the experiment and from three wether lambs from the College herd at the conclusion of feeding trial in the Spring of 1954. The determination of 17-ketosteroids in these samples by the Zimmerman reaction showed a marked increase in these compounds in the urine of the treated lambs. These results are in accord with the view that the enlargement of the male tissue is caused, in part at least, by hormones produced by the adrenal cortex.

BLOOD FAT LEVELS IN TWO BREEDS OF GILTS ON DIFFERENT FEEDING LEVEL SEQUENCES. H. L. Self, R. H. Grummer and L. E. Casida, *University of Wisconsin.*

Blood fat levels (by method of Allen) were studied in the Chester White (CW) and Poland China (PC) breeds on three different sequences of feeding levels: i.e., (FF)—self-fed from 70 days of age to second heat; (LL)—limited-fed (hand-fed 2/3 of self-fed consumption) from 70 days of age to second heat; (LF)—limited-fed from 70 days to puberty and then changed to self-fed. Whole blood samples were taken from each gilt at approximately 4½ to 5 hours after the AM feeding time on the 7th to 9th day after the beginning of her first estrous period. The 141 Mg% mean of the 30 CW gilts was higher ($P < 0.05$) than the 127 Mg% mean of the 32 PC gilts. The 128 Mg% mean of the FF gilts did not differ from the 125 Mg% mean of the LL gilts; however, the 155 Mg% mean of the LF gilts was higher ($P < 0.05$) than the means of either the FF or LL groups. There was no significant association within the different groups between the blood fat level and the average daily gain during the first estrous cycle ($r = 0.06$), nor between blood fat level and weight at the time the sample was taken ($r = 0.03$). However, blood fat level was significantly associated with the ovulation rate at second heat ($r = -0.30$; $P < 0.05$); as well as with age at the time the sample was taken, which is confounded with age at puberty ($r = 0.27$; $P < 0.05$).

THE RELATIONSHIP OF THYROID ACTIVITY TO LACTATION, GROWTH, AND SEX IN SHEEP. O. N. Singh, H. A. Henneman and E. P. Reineke, *Michigan State College.*

Using radioactive I^{131} the thyroid output turnover half-time was studied on 11 lactating ewes. The turnover half-time varied from 144.7 hours to 358.3 hours The coefficient of correlation between the output t ½ in 7 lactating ewes and the gain of their suckling twin lambs during the first 3 weeks of life was —0.552 ($P < .05$). This indicates that the lactating ewe with a greater thyroid activity probably secretes a larger quantity of milk, resulting in an increased growth of the lambs. There was no apparent relationship between the output t ½ in 4 lactating ewes and the gain of suckling single lambs. The thyroid secretion rate was also measured by the extrapolation technique using 12 ewe lambs, 5 ram lambs and 4 wether lambs, all of the Shropshire breed. The correlation coefficient between 1-thyroxine secretion of the five months old ewe lambs and a month's gain in weight was 0.792 ($P < .01$). The ewe lambs with the higher thyroid activity gained faster than the ewe lambs with a lower thyroid secretion. The average daily 1-thyroxine secretion rate per 100 lb. of body weight for ewe lambs was 0.082 mg.; for rams, 0.066 mg.; and for wethers 0.048 mg. The difference between the secretion rates of ewe and wether lambs was significant at the 5% level.

THE GROWTH AND SEXUAL DEVELOPMENT OF YOUNG HOLSTEIN HEIFERS AS INFLUENCED BY THREE LEVELS OF NUTRITION. A. M. Sorensen, Jr., R. W. Bratton, W. Hansel, W. H. Hough, *Cornell University.*

Sixty-four Holstein heifer calves were randomly allotted to three groups of twenty-one calves each and subjected to three different levels of nutrition in an attempt to measure the effect of nutrition on reproductive performance. The three levels of nutrition were based on Morrison's TDN standard requirements and designated as high level, 140%; medium level, 100%; and low level, 60%. Four calves from each group were slaughtered at 16 week intervals at which time

organ weights and sexual development were observed. Records of estrus and growth were maintained throughout the lives of the individuals. The calves on the high level of nutrition had reached an average age of 37.4 weeks at the time of the manifestation of signs of first estrus as compared to 47.1 weeks for calves on the medium level and 65 weeks for calves on the low level. The calves on the high level weighed an average of 579.5 lb.; medium, 596.6 lb. and low, 502.3 lb. at the time of first estrus. The high level calves had reached a greater height by the time of first estrus but a shorter length than the calves on the other two levels. The medium and low level calves showed very similar dimensions at this time. Udder measurements were made on all the animals during a period from three to five months of age. The level of nutrition apparently accounted for as much variance as did the genetical differences between animals.

THE EFFECT OF RAPID GROWTH WITH FATTENING UPON LACTATION IN CATTLE AND RATS. E. W. Swanson and T. R. Spann, *University of Tennessee.*

Identical twin grade Jersey heifers were divided at 3 mo. after which one group was fed concentrates *ad lib.* and the other was fed normal rations with no grain after 1 yr. The grain-fed heifers fattened and grew rapidly. At breeding age, 15 mo., the averages for super- and normal-growth groups, respectively, were 628 and 548 lb. weight, 61.0 and 57.4 in. heart girth, and 43.0 and 42.7 in. withers height. Just before parturition, 24 mo., they averaged 912 and 781 lb. weight, 67.5 and 63.2 in. heart girth, and 46.5 and 45.8 in. withers height. After parturition they were fed identical rations. Two pairs have completed two lactations. The fattened heifers failed to milk as long or as much as the normal heifers. In the 1st lactation the former averaged 767 lb. milk and 27.2 lb. fat in 106 days. The normal ones averaged 1345 lb. milk and 53.5 lb. of fat in 106 days. In the 2nd lactation the fattened ones averaged 768 lb. of milk and 28.9 lb. of fat in 68 days, while the normal ones averaged in the same time 1437 lb. of milk and 56.5 lb. of fat. A similar experiment with white rats was conducted in which lactation was measured by the growth of equalized litters, 11 young per rat. Rats raised by *ad lib.* feeding raised only 59% of their young to 21 days with an average litter gain of 136 gm. Rats fed 80% of *ad lib.* to parturition raised 93% of their young with an average litter gain of 235 gm. In the second lactation the rapid-growth rats produced only 60% as much as the restricted rats. The mammary glands of the fattened animals were not fully developed. These results indicate that excess fattening during growth is detrimental to lactating ability.

GROWTH OF NEWBORN PIGS IMPLANTED SUBCUTANEOUSLY WITH BACITRACIN PELLETS. Melvin J. Swenson, Ralph G. Buckner, Dennis D. Goetsch, C. E. Aubel and G. K. L. Underbjerg, *Kansas State College.*

Forty-seven newborn pigs from six gilts were randomized to three groups. One group was injected subcutaneously, posterior to the right ear, with a bacitracin pellet 36 hours after birth. Another group was injected with a zinc-aluminum-bacitracin pellet. The third group was left as a control. Growth of the nursing pigs was not altered by the implanted antibiotic pellets.

SOME EFFECTS OF ADDING SYNTHETIC ANDROGENS TO THE FATTENING RATION OF EWE LAMBS. Bruce Taylor, W. H. Hale and Wise Burroughs, *Iowa State College.*

Sixteen western ewe lambs averaging 69 lb. in weight were randomly allotted to four groups for an 84-day trial conducted in the December to March period

of 1953–1954. The lambs were individually fed an 11.5% protein basal ration of cracked corn, soybean oil meal, cane molasses and ground alfalfa hay. Additions to the basal ration consisted of: (1) none; (2) 2.41 mg. methyl testosterone per pound of ration; (3) 12.07 mg. methyl testosterone per pound of ration; and (4) testosterone propionate, in oil, injected subcutaneously each 14 days at the rate of 3.3 mg. per 100 lb. of body weight. Both methyl testosterone fed groups gained 16% more than the controls; whereas the testosterone propionate injected lambs gained 34% more. Feed efficiency favored the treatments, with group 4 requiring 19% less feed per unit of gain. Carcass grades and dressing percentages were highest in the group fed the higher level of methyl testosterone and lowest in the testosterone propionate injected lambs. Thyroid weights were heaviest in group 4. In a second experiment with 79 lb. ewe lambs, conducted in the period of March 31 to June 21, 1954, the same endocrine stimulants produced slightly but not statistically significantly greater gains. Seasonal differences may well affect stimulation of the ewe lamb with synthetic androgens.

SOME EFFECTS OF ANTERIOR PITUITARY GROWTH HORMONE ON SWINE. E. J. Turman and F. N. Andrews, *Purdue University*.

Three trials were conducted to study the effects of daily intramuscular injections of a purified bovine anterior pituitary growth hormone preparation in hogs. The pigs were started on experiment at approximately 100 lb., and carried to a final weight of approximately 220 lb. Data were obtained on seven experimental and seven control animals. There were no significant differences in daily gains, 2.26 lb. for the injected pigs and 2.19 lb. for controls. There were highly significant differences in feed efficiency, the injected pigs required 310 lb. of feed per 100 lb. gain while the control pigs required 384 lb. of feed. There were highly significant differences in carcass length and backfat thickness, 30.57 inches and 1.40 inches for injected pigs and 29.33 inches and 1.77 inches for the controls. The chemical composition of the carcasses of growth hormone injected and control pigs respectively were: protein, 13.5% and 10.8%; fat, 36.8% and 49.3%; and moisture, 49.1% and 39.4%. All differences in chemical composition were highly significant. Daily injections of growth hormone resulted in significant increases in blood glucose, and slight, but not significant, increases in blood levels of inorganic phosphorus and decreases in blood levels of non-protein nitrogen. The pituitaries from growth hormone injected pigs had a significantly greater concentration of thyrotropic and gonadotropic hormones than did pituitaries from the control pigs. Marked pathology was observed in some animals in Trials 2 and 3.

PREGNANCIES RESULTING IN A VARIABLE GROUP OF COWS AFTER INJECTIONS OF PROGESTERONE. L. C. Ulberg, Ralph Boulware, Richard E. Deese, and C. E. Lindley, *Mississippi State College*.

A total of 43 beef females was allotted according to one of the following reproductive classifications: (I) non-parous, 2-year-old; (II) non-parous 3-year-old; (III) parous dry cows and (IV) parous cows with calves. They received daily injections of either 50 or 25 mg. of progesterone for 14 days. Thirty-five cows, mostly with calves, were used as a check on inseminations. The treated animals were inseminated at the second post-treatment heat, with pregnancy diagnoses made 35–45 days later. The interval from last injection to second post-treatment heat in 19 animals on the 50 mg. dosage was 15–30 days (avg. 24.8) and for 18 animals on the 25 mg. dosage was 13–34 days (avg. 25.3). The number of pregnancies was 11/19 (57.9%) and 13/18 (72.2%) for the high and low dosages respectively. By breed, in the treated animals, 11/19 (57.9%) and 13/18

(72.2%) were pregnant while in the non-treated 7/11 (63.6%) and 16/24 (66.7%) were pregnant in Angus and Hereford cows respectively. By reproductive classifications 7/13 (53.8%); 5/8 (62.5%); 2/5 (40.0%) and 10/11 (90.9%) were pregnant in Lots I, II, III, and IV respectively. When arranged according to body weights at the end of treatment, pregnancies were 7/13 (53.8%), 11/18 (61.1%) and 6/7 (85.7%) for those under 900 lb., those 900–1099 lb. and those over 1100 lb. respectively. However, when animals weighing above the mean within each reproductive classification were pooled 15/17 (88.2%) were pregnant while those weighing less than the mean 9/19 (47.4%) were pregnant.

THE FREEZABILITY OF BULL SPERM AS AFFECTED BY INTERACTIONS OF GLYCEROL LEVELS, RATES OF FREEZING, AND SUBZERO STORAGE TEMPERATURES. N. L. VanDemark and W. C. Kinney, Jr., *University of Illinois.*

Studies have been carried out to determine the interactions between glycerol levels, citrate levels, rates of freezing, and subzero storage temperatures on the freezability of bull spermatozoa. A cooling rate of 2° C. per minute from +5 to −19° C. and 4° per minute from −19 to −79° C. resulted in higher sperm survival than cooling at a rate one-fourth as fast or 4 times as fast with glycerol levels of 3, 5, 7, 9, and 11% (final concentration) when the citrate level was held constant (1.55% final concentration). The detrimental effect on sperm survival of increasing or decreasing glycerol levels above or below 7% was more pronounced at citrate levels of 1.15% and 2.35% than at 1.75%. The fastest freezing rate was least injurious with the highest citrate and highest glycerol level combination. However, the highest glycerol level was the most injurious to unfrozen semen, and dilution immediately upon thawing to reduce rapidly the glycerol percentage was detrimental.

While the optimal glycerol level for storage of sperm at −20° C. was found to be approximately 19%, the percentage of surviving sperm was much lower than at a storage temperature of −79° C., and even this low percentage had decreased further when the storage period was lengthened by only one day.

EFFECT OF VARIOUS FACTORS UPON EXPERIMENTAL ERROR IN NONRETURN DATA FROM ARTIFICIAL INSEMINATION. E. L. Willett and J. I. Ohms, *American Foundation for the Study of Genetics.*

Sixty- to 90-day nonreturn rate data were studied from a field trial involving 96,515 first services from 1080 collections from 90 bulls and in which four treatments were compared: no antibacterial agents, sulfanilamide and streptomycin, streptomycin alone, and penicillin and streptomycin. When included in the yolk-citrate diluent, sulfanilamide was added to give a final concentration of 0.3%; streptomycin, 500 per ml.; and penicillin, 500 units per ml. Collections were not split, the four treatments were randomized in each block of four successive collections from each bull, and each bull was represented by three blocks. Experimental error was the mean square for collections treated alike within bulls. The data were divided by bulls into five class intervals based upon av. numbers of services per collection. The different class intervals, the number of collections represented in each, and the over-all error mean square for each were: 9–22, 324, 218; 29–56, 240, 90; 60–101, 216, 56; 147–183, 144, 47; 224–277, 156, 41. Within each of the two lowest interval groups there were no significant differences among the individual treatment error variances; within the other three groups the differences among error mean squares were significant, with those for no antibacterial agents being largest. The over-all error mean square for thirteen bulls

which were found to be infected with *Vibrio fetus* was close to the value expected with the average number of services per collection for these bulls.

HEATED HOMOGENIZED MILK VS. EGG-YOLK CITRATE AS A BULL SEMEN EXTENDER. James A. Williams, Richard W. Green, A. C. Baltzer, *Michigan State College.*

Heated homogenized milk was prepared according to the method of Thacker and Almquist. This product was compared to 1:1 egg-yolk citrate extender used by the Michigan Artificial Breeders Coop. The experiment involved a split sample technique. Only Holstein semen was used. Yolk citrate and milk extended semen was used alternately in the routine breeding of cows in four different locals during February and March 1954. Comparison of progressive motility of the spermatozoa in the split samples was made in the laboratory. No appreciable difference was noted. Accurate evaluation of motility was difficult in homogenized milk. Field results showed 543 first services performed with the milk extended semen with 70.9% non-return, based on 60–90 day returns. There were 527 egg-yolk first services with 65.1% non-return. The "t" test showed highly significant difference between the two percentages. One of the four locals found no difference. Another comparison was made for the first half of February when egg-yolk extended semen was used by all locals not on the split sample problem. This included all breeds in which 6558 first services were performed with 62.9% NR on 75–90 day return. During the last half of February, homogenized milk extended semen was used by the same locals, performing 6120 first services with 70.0% NR. The difference was highly significant. Eleven of the 31 bulls used on the split sample experiment, showed no improvement with the milk extended semen as based on their percent non-return.

DEVIATIONS FROM THE EXPECTED ESTROUS CYCLE BEHAVIOR OF PREGNANT AND NON-PREGNANT EWES. S. M. Williams, U. S. Garrigus, A. V. Nalbandov and H. W. Norton, *University of Illinois.*

Approximately 106 crossbred ewes were checked for heat daily throughout three consecutive years. A total of 2975 estrous cycles was observed of which 2322 lasted 14–19 days (normal cycles), 358 were longer than 19 days (long cycles) and 295 were shorter than 14 days (short cycles). Long and short cycles were characteristic of the beginning and end of the breeding season. The percentage of long cycles fell and that of short cycles rose as the age of the ewes increased. During the third breeding season the ewes were randomly divided into three groups as they first came in heat and they were bred at the 1st, 2nd, and 3rd heats respectively. Half of the bred ewes were slaughtered between two and seven days to determine ovulation and fertilization rates. The remainder were slaughtered between 20 and 30 days to determine embryonal mortality and fertility. All ewes were checked for heat until slaughtered. Twenty-two percent of ewes returned to service, their subsequent fertility and cycle lengths were comparable with those settled on first service. Eleven of the fifty pregnant ewes had 1–3 heats during gestation, the intervals between conception and the first heat being 22.4 ± 8.8 days. No significant difference existed between the ovarian characteristics (corpora lutea and follicles) and subsequent fertility of these ewes and the ewes which showed no heats during pregnancy. Ewes bred following a short or a long cycle had the same reproductive efficiency as ewes bred after cycles of normal length.

Teaching Section

F. N. Andrews, *Chairman*

LIVESTOCK IN THE MIDDLE EAST. L. J. Horlacher, *University of Kentucky.*

The richest and most influential peasant in a village is the man who owns the most cattle or sheep. Fat-tailed, coarse-wooled sheep are herded in summer on high mountain range and in winter on flat, parched desert. Some are fed barley while others receive no supplemental feed. All sheep must be housed within the village walls at night to prevent stealing and loss from predatory animals. Goats are produced in large numbers for milk and hair, but they destroy young trees and help keep pastures bare. Cattle have three functions: Meat, milk, and work. Swiss, French and other European breeds are used in artificial insemination program, with fair results. There is not enough feed to produce good beef. Horses are decreasing in number and are seldom used as work animals. Many tribes have good horses which are useful for transportation during migration. The armed forces use many horses and mules. Donkeys are the universal beasts of burden. They are cheap in price and consume little feed, therefore they are economical. The ship of the desert, the camel, is used for long-distance transportation, but trucks are tending to take over this job. The value of a camel is much higher than that of a donkey, but cost of maintenance is low. There is little interest in improving pastures and forage and the number of animals is definitely limited by the amount of feed available. No good books on livestock have been written for this area.

NEWS AND NOTES

The forty-sixth annual meeting of the American Society of Animal Production will be held in the Hotel Sherman in Chicago, Illinois on November 26 and 27, 1954, with a dinner for the honored guest on November 28. Abstracts of most of the papers which are to be presented at the meeting are published in this issue of the JOURNAL OF ANIMAL SCIENCE.

The nominating committee of the American Society of Animal Production composed of G. Bohstedt, H. M. Briggs and E. W. Crampton has proposed the following list of candidates to be presented at the next election of officers at the annual business meeting in Chicago on November 27:

For President: J. I. Miller.
For Vice President: L. E. Casida
H. H. Cole
For Secretary: W. M. Beeson

A. M. Pearson has recently moved from the University of Florida to Michigan State College where he will be engaged primarily in meats research in the Department of Animal Husbandry.

Artificial Breeding and Livestock Improvement by G. W. Stamm was published by Popular Mechanics Press, Chicago, in July, 1954. It has been written especially for the farmer in non-technical language.

James L. Carmon, who has been on leave of absence from the University of Georgia for the past two years completing the requirements for his PhD degree in Animal Genetics and Statistics at North Carolina State College returned to the former institution on a joint teaching and research assignment as of August 15, 1954.

The current organizational alignment for animal work of the Agricultural Research Service of the U.S.D.A. and principal personnel assignments are as follows:

Administrator, Agricultural Research Service—B. T. Shaw
Deputy Administrator for Research—George W. Irving, Jr.
Director, Livestock Research—O. E. Reed
Assistant Director, Livestock Research—Hugh C. McPhee
Chief, Animal Disease and Parasite Research Branch—B. T. Simms
Head, Bacterial and Mycotic Diseases Section—C. A. Manthei
Head, Viral and Rickettsial Diseases Section—L. O. Mott
Head, Non-Infectious Diseases Section—A. M. Lee
Head, Chemistry and Physics Section—H. W. Johnson
Head, Protozoan Parasite Section—L. A. Spindler
Head, Parasite Treatment Section—A. O. Foster
Head, Helminth Parasite Section—E. W. Price
Chief, Animal and Poultry Husbandry Research Branch—T. C. Byerly
Head, Cattle Research Section—L. L. Madsen
Head, Poultry Research Section—A. W. Brant
Head, Swine Research Section—John H. Zeller
Acting Head, Sheep, Goats and Fiber Section—D. A. Spencer
Head, Meat Production and Quality Research Section—N. R. Ellis
Chief, Dairy Husbandry Research Branch—Ralph E. Hodson
Head, Breeding, Feeding and Management Section—M. H. Fohrman
Head, Nutrition and Physiology Section—L. A. Moore
Head, Dairy Herd Improvement Sire-Proving Section—J. Frank Kendrick

The North Atlantic Branch of the American Society of Animal Production met at Pennsylvania State University, State College, Penna. on June 21, 1954. The new officers who were elected are as follows: President—Nathan S. Hale, University of Massachusetts; Vice President—Geo. W. Vander Noot, Rutgers University; Secretary-Treasurer—W. W. Green, Universty of Maryland. The 1955 meeting will be held at Cornell University.

W. L. Blizzard, well-known livestock judge and former Dean of the School of Agriculture and Director of the Agricultural Experiment Station at Oklahoma A. & M. College passed away on July 3. He had been ill following a stroke suffered in January, 1952. Dean Blizzard joined the Animal Husbandry Department staff in 1915 and was made head of that department in 1939.

G. H. Hart, Dean of the School of Veterinary Medicine at the University of California, retired on July 1.

L. A. Maynard, Head of the Department of Biochemistry and Nutrition at Cornell University recently received the $1000 Osborne and Mendel award for his fundamental research on the biochemical and nutritional aspects of lipid metabolism and of lactation and for his contributions as teacher, administrator, and public servant in the field of nutrition.

K. F. Warner, who served for many years as extension meat specialist in the U. S. Department of Agriculture, was awarded the honorary degree of Doctor of Agriculture by the University of Nebraska on June 7. He is now on the staff of the Foreign Agricultural Service of the Department.

W. M. Mohler, a member of the Animal Disease and Parasite Research Branch of the U. S. Department of Agriculture died of a heart attack on June 9. He was the son of John R. Mohler, for many years Chief of the Bureau of Animal Industry.

D. W. Colvard, Dean of the School of Agriculture at North Carolina State College, and former Head of the Animal Industry Department at that institution, was recently appointed to the Advisory Committee of the Kellogg Foundations' Division of Agriculture.

H. R. Purdy, who was formerly instructor in animal husbandry and coach of livestock judging at Ohio State University, has joined the Department of Animal Science staff at Pennsylvania State University, in charge of beef cattle.

G. R. Wilson, former County Agent in Butler County, Ohio, is now in charge of beef cattle work at Ohio State University. He succeeds Herman Purdy.

A. H. Jensen, a former member of the Animal Science Department of Iowa State College, is now Assistant Professor of Animal Husbandry at the University of Illinois.

J. M. Vial, well-known extension animal husbandman at the University of Maryland, retired from this position on July 1, 1954. He will become manager of the Eastern National Livestock Show at Baltimore.

Carl B. Roubicek formerly regional coordinator for the Western Regional Beef Cattle Breeding Project has been appointed professor of Animal Husbandry and director of research in the Department of Animal Industry at the University of Arizona.

O. M. Nelson, professor of Animal Husbandry at Oregon State College retired June 30, 1954.

H. L. Self who recently received his doctorate in animal physiology at the University of Wisconsin has joined the staff of that institution as assistant professor.

E. E. Goodwin is now assistant professor and assistant animal husbandman at Washington State College, Pullman, Wash.

V. H. Arthaud has been transferred from the beef cattle research station, Front Royal, Va., to the University of Nebraska where he will be engaged in research on dwarfism.

T. J. Marlowe has been made associate professor of animal husbandry at the Virginia Agricultural Experiment Station, Blacksburg.

D. C. Meyerhoeffer has been appointed instructor in animal husbandry at Virginia Polytechnic Institute, and is located at the Front Royal station.

D. L. Handlin has been made assistant professor in the Animal Husbandry Department at South Carolina State College.

R. J. Deans has been appointed instructor in the Animal Husbandry Department at Michigan State College.

Eugene Birmingham, Charles J. Heidenrich, H. B. Hedrick and Granville B. Thompson are new members of the animal husbandry department at the University of Missouri with the rank of instructor.

W. T. Crandall, professor of Animal Husbandry at Cornell University, retired July 1.

H. D. Naumann has joined the Animal Husbandry Department at Cornell University as assistant professor.

W. G. Black is now associate professor of animal husbandry at the Massachusetts Agricultural Experiment Station. He was formerly with the Animal Husbandry Department at the University of Nevada.

Howard G. Weeth, who was formerly at the University of Hawaii, has been appointed animal physiologist at the University of Nevada College of Agriculture. He succeeds W. G. Black.

L. N. Baker and J. S. Greenstein are new members of the Animal Husbandry Department at the University of Massachusetts with the rank of assistant professor.

S. H. Fowler, who recently completed his doctorate at the Texas A & M College has been appointed assistant professor of Animal Husbandry at the University of Maryland.

D. E. Jasper has been appointed chairman of the Department of Veterinary Science at the University of California at Davis. He succeeds George H. Hart who retired recently.

William F. Brannon has joined the staff of the Squaw Butte-Harvey branch experiment station at Burns, Oregon, where he will be in charge of livestock research. He received his Ph.D. in animal nutrition at Cornell in 1953 and has been extension animal husbandman at that institution during the past year.

Recent additions to the animal husbandry staff at South Dakota State College include L. F. Bush, associate professor and associate animal husbandman, and L. E. DuBose, assistant professor and assistant animal husbandman.

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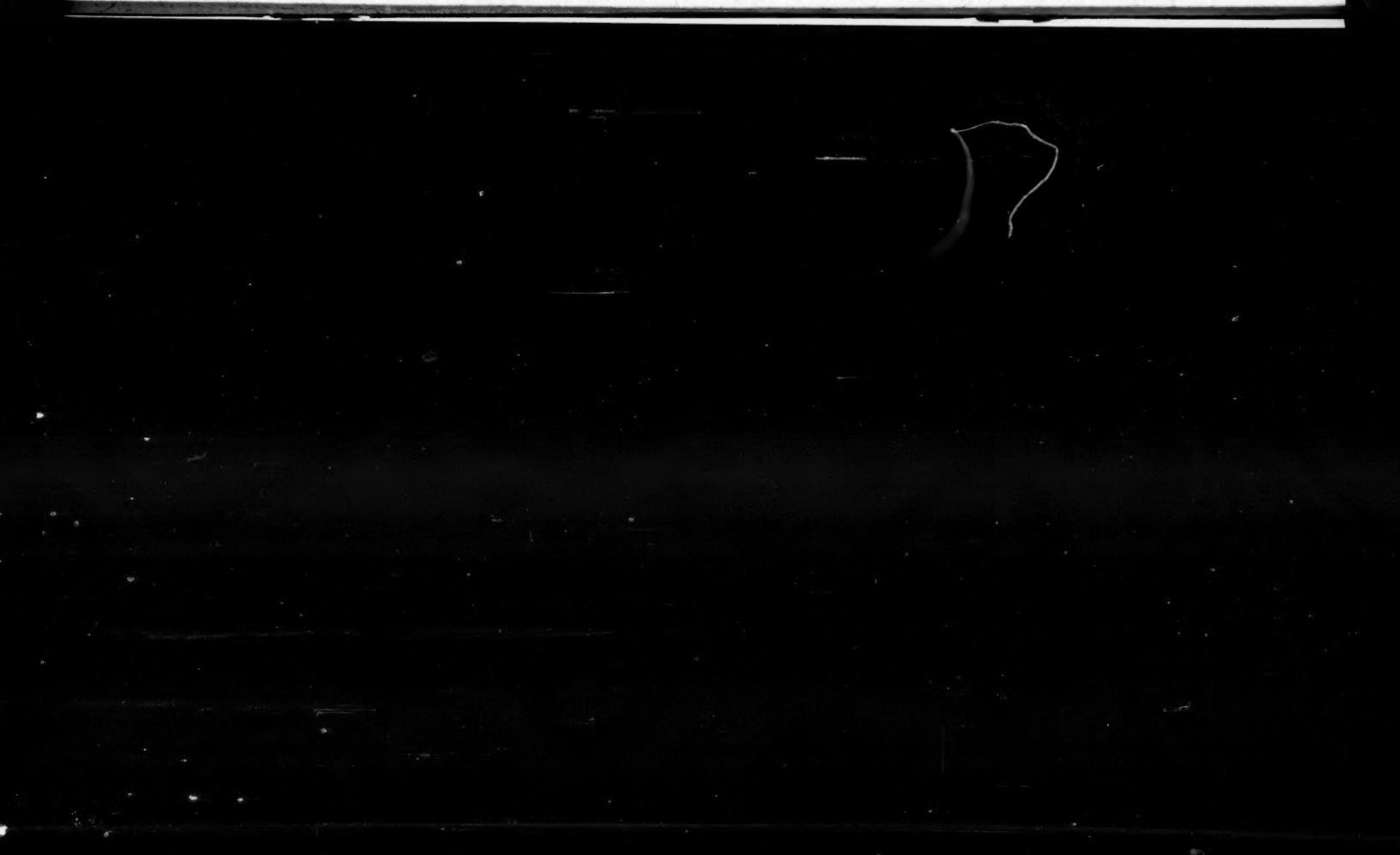

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1. Papers may be submitted reporting work whose ultimate objective is clearly the solution of a livestock production problem and whose contents should be of interest particularly to those dealing with livestock production problems. Review papers on subjects of general interest will also be published at intervals, and will be prepared on invitation by the Editorial Board.

2. All papers will be subject to critical review by the Editorial Board, or others designated by the Editor. Papers needing revision will be returned to authors and should be revised and returned promptly. Papers not suitable for publication will be returned to authors with a statement of reasons for not accepting them.

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   (c) Tables should be as few and as simple as is feasible for presentation of the essential data.

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8. A "News and Notes" section will be published in each issue, including Society announcements, changes in positions and other items of professional interest concerning the members, and other items of interest to the membership. All such items should be sent promptly to the Editor.

9. Abstracts of papers to be presented at the annual meetings of the Society will be published in the November issue. Such abstracts must not exceed 250 words in length, should be typed (double spaced) and the ribbon copy submitted, the lines on the page should be numbered, in the left margin, from top to bottom, and must be in the hands of the Secretary of the Society by August 15, preceding the meeting. These abstracts must be summaries of results and conclusions.

10. Presentation of a paper at the annual meeting does not exclude it from publication in complete form in the JOURNAL. Such papers may be submitted to the Editor and will be given equal consideration with others submitted.

11. All manuscripts and correspondence concerning them should be addressed to the Editor: Dr. L. E. Casida, Department of Genetics, University of Wisconsin, Madison, Wisconsin, or to the publishers: Boyd Printing Company, 372–374 Broadway, Albany 7, New York.

12. All correspondence concerning subscriptions to the JOURNAL OF ANIMAL SCIENCE and other business matters should be addressed to the Business Manager, Dr. W. M. Beeson, Department of Animal Husbandry, Purdue University, Lafayette, Ind., or to the publishers, Boyd Printing Company, 372–374 Broadway, Albany 7, New York.